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INTRANUCLEAR INCLUSIONS IN VISCERAL DISEASE *

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The presence of intranuclear bodies with specific tinctorial properties has been widely accepted as indicative of infection with a virus having special affinity for tissues of neural or ectodermal origin. It is the purpose of this paper to present morphological evidence that similar bodies may be found in lesions of internal organs, and to bring forward for consideration the possibility that viruses of this type may localize in tissues other than skin and central nervous system.

REVIEW OF LITERATURE

The history of such nuclear inclusions in mammalian tissue cells dates back to a publication by Jesionek and Kiolemenoglou [1] in 1904, on the finding of protozoön-like structures in the organs of an hereditary syphilitic. Their illustrations and descriptions show that they were observing bodies very similar to, or identical with, those with which this paper is concerned. They were found in the kidneys, lungs and liver of an eight-month syphilitic stillborn fetus. In the kidneys the cells containing them were irregularly scattered through the interstitial tissue of the cortex, often in groups of ten to forty. In the liver and lungs they occurred singly; in the lung they were found free in the bronchi and alveoli.

The intranuclear bodies are described as oval in form, with a fairly definite cuticular zone suggesting a capsule. The nucleus consisted of a central body, separated as if by a shell from the cytoplasm; on

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the inner surface of this shell (*i.e.* the nuclear membrane) were darkly stained granules of various sizes. The cell body was spongy and the pole near the nucleus filled with granules. As regards staining, the authors noted that the central body, that is to say the inclusion, stained rather palely and with a reddish tinge in hematoxylin-eosin preparations, after sublimate fixation, whereas the peripheral granules were dark blue.

The authors believed that they could exclude the possibility that these bodies arose as modifications from preformed structures. Their appearance suggested some animal or vegetable parasite, and after consultation with Professor R. Hertwig, it was concluded that they were probably Gregarines. The possibility that they were the cause of syphilis — this was before the days of the *Treponema pallidum* — was discussed, but not seriously entertained.

Shortly after the appearance of this paper Ribbert,[2] recalling that he had seen similar bodies some twenty years previously, published a short paper on protozoön-like cells in the kidney of a syphilitic newborn and in the parotid of children. In the kidneys Ribbert found large cells in the tubules but not in the glomeruli or stroma. The preparations were twenty years old and somewhat faded, but it was still evident that there were no transitions between these cells and the renal epithelium. They are described as having a round or oval nucleus containing a large homogeneous body, separated from the nuclear membrane by a clear zone. The cell body was slightly vacuolated or finely granular.

In Ribbert's second case similar intranuclear bodies were found in the parotid of a non-syphilitic year-old child. They occurred in the ducts singly or in groups, often pushing aside the epithelial cells. In his third case identical cells were found in even greater numbers in the parotid ducts.

As regards their significance Ribbert agreed with Jesionek and Kiolemenoglou that they were elements which in no way resembled any of the normal or pathologically altered body cells. This suggested at once their parasitic nature and the preparations were submitted to the zoölogists Rhumbler and Ehlers, who rather inclined to the opinion that they were amoebae or sporozoa.

In 1907, Loewenstein,[3] at Ribbert's suggestion, studied the parotids of thirty children from two months to two years old and found inclusions in four. In the last case they were observed in fresh un-

stained preparations. The slides were submitted to Professor Ludwig, the Director of the Zoölogical Institute, who was of the opinion that they were protozoa, either coccidia or other sporozoa. Loewenstein found no transitions to normal body cells.

Pisano,[4] in 1910, reported the finding of similar intranuclear bodies in the tissues of a stillborn fetus. The viscera in this case showed pronounced lesions — the liver, a gummatous hepatitis, while the kidneys, spleen, thyroid and lungs were all the seat of interstitial fibrosis. The large cellular elements containing inclusions were present in great number in the kidneys, fairly numerous in the liver and rare in the lungs. They were found in the crevices of the connective tissue and free in the lumina of the tubuli contorti of the kidneys. As to their significance various possibilities were considered and rejected. The most specious hypothesis, and the one adopted by Pisano, and also by Perrando [5] in a subsequent article, was that the cells were of epithelial origin but "arrested in their development by the dystropic and paraplastic effect of the syphilitic infection." The paper makes no reference to previous German observations. The following year Mouchet[6] found similar bodies in the bile ducts of an eight-day syphilitic infant, with icterus. They were regarded by him as sporozoa, the sporocysts of which were enclosed within hypertrophic epithelial cells.

Smith and Weidman [7] in 1910 published in the University of Pennsylvania Medical Bulletin a paper entitled "Infection of a Still-born Infant by an Amebiform Protozoan (Entameba mortinatalium), N. S." The bodies which they depicted and described were obviously identical in nature with those reported by Jesionek and Kiolemenoglou, and by Ribbert. One other fact of interest is brought out in their paper, that in the liver and kidney the supposed parasites were surrounded by a definite inflammatory reaction of lymphoid and polymorphonuclear leucocytes. In the light of subsequent events it is perhaps unnecessary to review the detailed steps by which Smith and Weidman excluded all the hitherto known amoebae as a result of which they were forced to create a new species for their parasite.

In 1914, the same authors,[8] having in the meantime become acquainted with the previous German reports, published a new observation. The case was that of a two-months' old child, dying of pneumonia. There were ulcerous eruptions about the mouth and nose and scaly lesions of the buttocks, no definite luetic parental

history, a negative Wassermann reaction, but fibrosis of thymus and of pancreas; so that there was a strong suspicion that the child, like the previous cases, was syphilitic. The "parasites," having the same appearance as those in the previous case, were found this time only in the lungs. There was an organizing pneumonia, and a few areas of caseous necrosis in which no tubercle bacilli could be found. The writers conclude that the "parasites" are "doubtless harmless for the mother, but for the fetus, especially when impaired by luetic taint, they may prove pathogenic and capable of destroying life."

Bodies of this type next reappear in the literature in 1921, being described this time as "An Intracellular Protozoan Parasite of the Duct of the Salivary Glands of the Guinea-pig." Leila Jackson [9] found it in 26 of 48 pigs examined, and described it as follows: "an encysted organism of irregular round or oval contour. In its most conspicuous and fully developed form, it practically replaces the host cell which still retains its relations to the duct wall. The center is occupied by a round or oval body, which stains deeply and unevenly, around which is a wide, lightly stained zone and outside of this a peripheral capsule." The illustrations which accompany the paper show a very close resemblance to the bodies described by the previous writers. Jackson, however, interpreted them as coccidial in nature, and thought they were situated within the cytoplasm of the epithelial cells, rather than within the nucleus. There may therefore perhaps be some question as to whether she was dealing with the same type of structure.

In the same year, 1921, Goodpasture and Talbot [10] published an excellent paper, "Concerning the Nature of the Protozoan-like Cells in Certain Lesions of Infancy." In a six-weeks' old male child, which had had green stools from birth, glucose in the urine, edema of the feet, and cough, they found large cells with acidophilic intranuclear bodies, in the alveoli of the lungs, in the chronically inflamed bronchi, and impacted in the glomeruli of the kidneys. Their illustrations leave no doubt that the bodies were identical with those in our own case, and with the bodies reported previously.

Goodpasture and Talbot remark that these inclusion-containing cells resemble no normal element of the body. They believe, however, that their origin can be traced to large mononuclear cells situated just outside the endothelium of small veins and capillaries, and that they may gain entrance into the blood stream, penetrating

the capillary walls. No cells of this type were observed in organs other than the kidneys, lungs and liver. The possibility that other types of cells, particularly epithelium, might be transformed into cells of this type, could not be excluded. Indeed, early stages of the transformation could be seen in the alveolar epithelial cells.

The authors found it difficult to form an opinion as to the nature of this remarkable change, which suggested the intranuclear bodies described by Tyzzer [11] (1906) in varicella. They felt very certain that whatever they might be, they were not protozoa.

De Lange [12] in 1922 recorded a similar finding in a three-day old icteric infant with cirrhosis of the liver. Syphilitic infection could not be proved, the viscera showing no spirochaetae by the Levaditi method, and the parents giving a negative Wassermann reaction. The inclusion-containing cells were found in the convoluted tubules of the kidneys; in their vicinity, there was round cell infiltration of the stroma. They were interpreted as some undetermined form of cell degeneration.

Three further cases were reported in 1922 by J. Müller,[13] one in a stillbirth, one in a child eight weeks old with hydrocephalus and slight interstitial nephritis, and a third in a two-months' old child with congenital syphilis. The inclusions were found only in the kidneys; their appearance conforms to that described by previous observers.

Müller excludes the possibility that the cells are protozoan because of their occurrence in stillbirths, and because it does not seem possible that they could have passed the placental barrier. He is therefore forced to believe that they originate from tissue cells which have undergone a peculiar degeneration, characterized by the dissociation of the oxy- and basi-chromatin within the nucleus, and by hypertrophy of the affected cells as a whole. The foregoing cases are summarized in Table 1.

We should hardly be justified in reviewing these isolated findings at length were it not for the fact that the matter of these intranuclear bodies takes on new interest and importance with the discovery of B. Lipschütz [14] in 1921 that similar structures are constantly and characteristically associated with the lesions produced by the herpes virus, both in man and rabbits. Although it had been previously shown by Grüter [15] and Loewenstein [16], that the virus of herpes febrilis is transmissible in series to rabbits, Lipschütz was the

first to make a careful study of the intracellular inclusions in this disease, and in herpes zoster and herpes genitalis, and to interpret these bodies as the expression or result of an intranuclear virus. Since Lipschütz's first publication, numerous articles have appeared, dealing not only with the question of cell inclusions, but with the most interesting and still obscure problem of the relation of the herpetic virus, and the encephalitis produced by certain strains of it in rabbits, to the virus of epidemic encephalitis in man. We shall attempt to discuss only the herpetic inclusions, and their possible bearing on the interpretation of the case which is here reported.

Lipschütz showed first of all that these bodies are not artifacts, inasmuch as they may be easily recognized in fresh preparations. They are round, oval, or even slightly irregular structures ranging from 2 micra to such as completely fill the nucleus, reaching to the nuclear membrane but leaving usually a small clear zone. Smaller inclusions lie against a clear background. Usually there is only one within a nuclear membrane, but sometimes several. Practically every nucleus at the site of the herpetic lesions may contain an inclusion. Frequently the inclusions appear homogeneous, but in properly differentiated iron-hematoxylin preparations, they appear to be composed of numerous minute granules embedded in a homogeneous matrix.

Although they typically occur within the nuclei, they may in early lesions be occasionally found in the cytoplasm in the vicinity of the nuclear membrane. They are present both in the nuclei of epithelial cells, and in the swollen hydropic nuclei of connective tissue cells in the corium, and in the cells about blood vessels. Once an inclusion was seen within a mast cell, and once within the proliferating epithelium of a hair follicle.

The staining reactions are summarized as follows:

Stain	Inclusions	Nucleoli
Giemsa	Red	Dark blue
Hematoxylin-eosin	Dark red	Blue-black
Heidenhain iron-hematoxylin	Yellow-gray	Black
Pappenheim	Green or blue	Red

The inclusions may thus be readily differentiated tinctorially from the nucleoli.

Lipschütz [14] discusses at length in this and a subsequent paper [17] the possible interpretation to be placed upon these and similar

TABLE 1

Résumé of Reported Cases

No.	Year	Author	Age	Pathological diagnosis	Location of inclusions	Interpretation
1	1904	Jesionek and Kiolemenoglou	Stillbirth	Congenital syphilis	Kidneys, lungs, and liver	Gregarines (R. Hertwig)
2	1904	Ribbert	Newborn	Congenital syphilis	Kidneys	Amoebae or Sporozoa? (Ehlers — Rhumbler)
3	1904	Ribbert	One year	——	Parotids	
4 5 6 7	1907	Loewenstein	Two months to two years	——	Parotids	Coccidia or other sporozoa (Ludwig)
8	1910	Pisano	Stillbirth	Congenital syphilis, Gummatous hepatitis, etc.	Kidneys, liver and lungs	Embryonic epithelial cells
9	1910	Mouchet	Eight days	Congenital syphilis	Bile ducts	Sporozoa
10	1910	Smith and Weidman	Stillbirth	Focal nephritis	Kidneys, liver, and lung	Entamoeba mortinatalium
11	1914	Smith and Weidman	Two months	Pneumonia	Lungs	Entamoeba mortinatalium
12	1921	Goodpasture and Talbot	Six weeks	Green stools, edema of feet, bronchitis	Lungs and kidneys	Degenerative change in the nuclei of endothelial leucocytes
13	1922	De Lange	Eight days	Congenital syphilis? icterus, cirrhosis	Kidneys	Undetermined form of cellular degeneration
14	1922	J. Müller	Eight weeks	Hydrocephalus, focal interstitial nephritis	Kidneys	Degenerative change, with dissociation of oxy- and basi-chromatin
15	1922	J. Müller	Stillbirth	?	Kidneys	
16	1922	J. Müller	Two months		Kidneys	

intranuclear inclusions, and reaches the conclusion that they represent the reaction of the nuclear plasm to a living virus classed with the Chlamydozoa-Strongyloplasma. To the group of these intranuclear viruses, which includes causative agents of the various forms of herpes, Borna's disease of horses (epidemic encephalomyelitis), varicella, fowl-pox, etc., is given the name "karyooikon group," distinguishing them from the cytooikon group, in which the inclusion is present in the cytoplasm. The trachoma bodies, the Guarneri corpuscle in smallpox, and the Negri bodies in the ganglion cells in rabies are representative of this latter group.

Lipschütz's arguments in support of the view that the inclusions are to be interpreted as a specific reaction to a living parasitic virus, are briefly the following:

1. Their general resemblance to the well-known inclusions of variola and vaccinia.
2. Their constant occurrence in the pathologic tissue, and their limitation to that tissue.
3. The impossibility of identifying them with hypertropic nucleoli or any known degenerative products of the cell.
4. The possibility of reproducing similar bodies by experimental inoculation of the virus.

Subsequent investigation has almost unanimously confirmed Lipschütz's work so far as the regular occurrence of the intranuclear bodies in spontaneous and experimental herpes lesions is concerned. There have, however, been differences of interpretation. Thus Lauda,[18] who studied the inclusions both in the herpetic lesion and within the ganglion cells and glia cells of the encephalitic rabbits, believes that the inclusions are the result of a degenerative, destructive process of the nucleus, whereby the oxychromatin comes to lie in the center of the cell, and the basi-chromatin moves to the periphery. Against the parasitic nature of the inclusions, he brings the following arguments: (1) the absence of evidence that the virus is really situated within the nucleus; (2) the absence of elementary bodies within the inclusions (he was unable to confirm Lipschütz's observation as to the presence of minute granules within the inclusion); (3) the lack of specificity, since bodies of apparently identical morphology are found in such diverse conditions as herpes labialis, zoster and varicella. Zdansky,[19] also, in a recent paper on

the pathologic anatomy of the encephalitis produced in rabbits by the herpes-encephalitis virus, ascribes no etiological importance to the inclusions found in the ganglion cells and glia cells, but regards them rather as degeneration products of the karyoplasma. Levaditi[20] and Da Fano [21] believe that the virus is more probably to be found in certain minute granular bodies scattered through the affected nerve tissue, both within the cells or between them. The larger intranuclear bodies (the "neurocorpuscules" of Levaditi, Harvier and Nicolau [22]) are regarded as products of nucleolar degeneration. Cowdry and Nicholson,[23] in studying cytologically experimental herpetic encephalitis material given them by Flexner, Noguchi and Amoss, attribute little importance to intranuclear inclusions of the Lipschütz type, and most of their attention appears to have been directed to the smaller types of granules emphasized by Da Fano. In conclusion, they state their belief that the inclusions which are so abundant in herpes do not represent a concrete class of granulations *sui generis*, but that they are of variable composition and derived from several sources.

Quite the opposite point of view is taken by Goodpasture and Teague. In one of the most recent of the brilliant series of studies in this field, Goodpasture [24] makes a very strong argument in favor of the view that the nuclear inclusions represent a change brought about specifically by the virus of herpes or closely related viruses. As the virus passes from the peripheral nerve endings to the central nervous system along the nerve trunks, a progressive involvement of the neurolemma cells and then of the ganglion cells in the corresponding region of the brain, may be demonstrated. When the lesion is unilateral, only the cells on the affected side show inclusions. While it is not maintained by Goodpasture that the intranuclear inclusion is itself the active agent of the disease, he does believe with Lipschütz that it indicates an intranuclear localization of the infective substance.

Case Report

F. S. (history 60991 — autopsy 9582), male, white, age 36 years. Entered Presbyterian Hospital September 27, 1924.

Chief Complaints. Fever for 3½ weeks. Pain in chest for 10 days.

Past History. Gonorrhoea 9 years ago. In 1908, an acute illness (appendicitis ?) since which time he has been subject to attacks of indigestion, characterized by acute pain across the upper abdomen and lower chest, coming on ½ hour after meals, and relieved by rhubarb and soda. At times the pain has been

localized in the right lower costal region and is relieved by lying on the affected side. No history of herpes was obtained on careful inquiry.

Present Illness. The present illness began gradually about one month ago with afternoon fever. Seven days after the onset a motile gram-negative bacillus was obtained from blood culture. This was at first believed to be *B. paratyphosus* but was later identified as *B. coli*. Ten days ago he began to have pains in the lower anterior part of the chest. The pain was burning in character and worst when fever was highest. There were no abdominal pains and no blood in the stools.

Physical Examination. Thin, well-developed man, acutely ill. Heart and lungs negative. Abdomen flat, with slight fullness in the right upper quadrant where there was definitely increased resistance and tenderness on pressure. Liver palpable 3 cm. below the costal margin. No abnormal neurological signs. Pulse regular, not dicrotic, 100. Blood pressure 105/55. Blood: R. B. C. 4,000,000, Hg. 92 per cent, W. B. C. 16,900, P. M. N. 79 per cent, Lym. 20 per cent, Eosin. 1 per cent. Wassermann reaction negative. Stool negative for blood (Guaiac test). Blood culture: negative on one occasion; on another, *B. coli* was again recovered.

Course. The temperature curve suggested a pyogenic infection rather than paratyphoid fever. The tenderness in the region of the liver persisted, and the diagnosis lay between subphrenic and hepatic abscess. On October 9th, exploratory coeliotomy disclosed an abscess in the right lobe of the liver, which was drained. Culture of the pus yielded *Staphylococcus pyogenes albus*.

Operation was followed by only temporary improvement. Transfusion (750 c.c. of blood) failed to influence the temperature or leucocytosis. Examination of the chest was negative.

On November 4th, because of the failure to improve, he was again operated upon, and a subphrenic abscess found and drained. The septic temperature persisted. An X-ray on November 8th showed a shadow in the right lower chest and obliteration of the phrenic angle. Dullness was found over this area. The leucocyte count was 24,850, P. M. N. 86 per cent.

Upon aspiration of the chest, a small amount of blood and fluid, sterile on culture, was obtained. On a second aspiration of the chest 10 days later, a small amount of blood-streaked pus was withdrawn, from which was grown a non-hemolytic streptococcus in pure culture. The margins of the draining operative wound at this time had become necrotic over a wide area, and masses of necrotic liver tissue were discharged from the abscess.

On November 19th, a piece of tissue was removed from the wall of the liver abscess. Sections of this showed bile ducts, some compressed, some distended with polymorphonuclears. There was much dense granulation tissue in the midst of which were a few degenerating liver cells; also many phagocytes, some containing hemosiderin. No amoebae could be found. No organisms could be found in a Levaditi preparation. Smears from the abscess stained with methylene blue, and by Fontana method, showed many cocci, often in chains, and many bacilli but no spirochaetae or spirillae, or filamentous organisms.

On December 1st, the patient developed bloody diarrhea and on December 5th *amoeba histolytica* was reported as present in the stools. The administration of emetin hydrochloride (0.63 gm. in twelve days) was followed by a decrease in the number of the stools. No amoebae were found after the emetin therapy was begun. The margins of the liver wound showed signs of more active

healing, with the appearance of granulation tissue, and the abscess cavity became smaller. The fever, however, persisted, and also the pulmonary signs.

On December 18th he had two bloody stools, the second one consisting of almost 500 c.c. of blood. He became weaker, and died two days later, having been ill approximately 3½ months.

Autopsy 9582. Anatomic diagnoses: Abscess of the liver; ulcerative colitis with hemorrhage; suppurative pleurisy, right; lobular pneumonia, organizing; small bronchiectases; fibrous pleural and peritoneal adhesions; sclerosis of pulmonary venules.

The body is that of a fairly well-developed, moderately emaciated white man, 165 cm. in length. There are no cutaneous lesions. There is rigor of the jaw, neck and extremities, and some lividity of the dependent portions. There are no abnormalities over the calvarium; no discharge from the eyes, ears, nose or mouth. The pupils are round, equal and in mid-dilatation. The conjunctivae and buccal mucous membranes are quite pale. The teeth are in good condition. There is no enlargement of the thyroid; the superficial lymph glands are not palpable. The chest is long and narrow. On the right side of the chest, in the posterior axillary line, over the 8th rib, is a recent surgical incision, through which a finger can be introduced directly into the pleural cavity. From this wound there flows a small amount of thick, milky-white, purulent material. There is a long incision over the lower anterior chest on the right, following the course of the ribs. The cartilages of the lower ribs project into the wound and are necrotic. There is considerable erosion of the epithelium around the margin of this wound, especially at the inner angle, where there is a granulating surface 2 cm. in breadth, extending back from the edge. The wound is widely gaping, and through it one can see directly into a cavity which extends into the liver. In the upper right quadrant of the abdomen are two gray, glistening scars which touch each other at their upper ends and form an angle of about 60°. The external genitalia are normal. There is no edema of the feet or ankles. The subcutaneous fat is practically absent.

Abdominal Cavity. There is no excess of fluid in the peritoneal cavity. There are fairly recent adhesions between the almost fat-free omentum and the anterior abdominal wall, and also in the region of the cecum. These adhesions become denser in the region of the liver and the anterior surface of the right lobe is bound to the anterior abdominal wall by tough adhesions which completely wall off the abscess from the peritoneal cavity. The liver edge extends 9 cm. below the xiphoid.

The *liver* weighs 1480 grams and measures 20 x 20 x 10 cm. After dissecting off the adhesions, there is seen in the right lobe on the anterior surface a cavity with an opening about 2 cm. in its greatest diameter. The surface of the right lobe of the liver is for the most part devoid of adhesions except in the region of the cavity; the left lobe is covered with a smooth glistening capsule. There is no accumulation of pus between the liver and diaphragm. On section it is seen that the cavity extends into the liver for a distance of 3 cm. The wall is composed of gray, glistening, dense fibrous tissue, superficially bile-stained and covered with a little exudate. Above the cavity, which is in the dome of the liver, there is an infarct-like area of atrophy and congestion, slightly sunken below the level of the surrounding parenchyma. This wedge-shaped area has its base toward the capsule and its apex is against a large branch of the hepatic vein. Contrasting with this, the lobules in other portions of the liver are considerably

larger; there is a very narrow red zone about the efferent vein, but about the portal vessels is a broad grayish-yellow zone. Surrounding the abscess cavity and extending for a variable distance toward the inferior border on the anterior surface is again seen an irregular zone, identical in appearance with the large one described. A communication between the larger bile ducts and the abscess cavity cannot be demonstrated, but there are seen in the wall of the cavity several oval openings from 1 to 2 mm. in diameter, which appear to be the openings of bile ducts. The portal vessels and bile ducts elsewhere appear normal. There are no other abscesses or infarcted areas found. There is no general increase in connective tissue.

The *gallbladder* is small; its wall is thin. It contains a small amount of pale viscid bile.

The *spleen* weighs 225 grams and measures 16 x 7 x 3 cm. The capsule is smooth and glistening. The organ is moderately soft. On section the pulp is reddish-gray in color; the Malpighian bodies are small but easily seen.

The *pancreas, adrenals, kidneys,* and *pelvic organs* are normal.

Gastro-intestinal tract. The stomach, duodenum, jejunum and ileum are normal.

In the cecum are found discreet ulcers of varying size with clean bases; only a few of these show slight undermining. These ulcers extend down to the inner layer of muscle and tend to encircle the gut; they measure 0.5 cm. to 4 cm. in greatest dimension. The wall about them is not indurated, and clinging to the margin of one of these ulcers are blood clots. In the ascending colon also are a few ulcers, to one of which a blood clot is adherent. They are similar in appearance to those in the cecum, but somewhat smaller. The bases of the ulcers are covered neither by exudate nor by definitely recognizable granulation tissue. There are fifteen of these ulcers. No other ulcers are found in the remainder of the large intestine.

The *lymph glands* in the mesentery are not enlarged; on section they appear normal.

Thoracic cavity. There are numerous adhesions between the lower lobe of the right lung and the parietal pleura. These adhesions tend to wall off the small cavity communicating with the incision in the lateral chest wall. A few adhesions are present between the apex of the upper lobe on the left and the parietal pleura. There is no excess of fluid in either pleural cavity.

The upper lobe of the *right lung* is everywhere air-containing and fluffy, as is also the middle lobe. The lower lobe is much collapsed, dark red and flabby; its pleura roughened by the fibrous adhesions and by a fibrino-purulent exudate. On section the cut surface of the upper and middle lobes is pale. There is no consolidation. The lower lobe on section is dark red, firm, but not uniformly consolidated. The pleura is quite obviously thickened; it seems also somewhat wrinkled, yet the exudate fills in the spaces between these wrinkles, producing a level surface. Scattered throughout the lower lobe are small grayish areas which appear to be more or less confined to the immediate region of the smaller bronchi. The bronchi are not thick walled.

The upper lobe of the *left lung* is everywhere air-containing. The lower lobe is heavy. The posterior portion is dark bluish-red in color, and this area is somewhat sunken below the level of the air-containing lung adjacent to it. The lower lobe feels flabby; it does not have the consistence of a firmly consolidated lung but is more or less elastic. On section there are found in the

upper lobe a few areas, lobular in size, which are consolidated, and the cut surface of these is yellowish in color. In the lower lobe are small areas of consolidation which are for the most part close to the smaller bronchi; they rarely measure more than 0.5 cm. in diameter, and are not confluent. The bronchi in the centers of these in some instances contain a little purulent fluid, but for the most part they are empty and their walls are not especially thickened. Some of the medium-sized bronchi are dilated; they are, however, lined with smooth mucosa. The lymph glands at the hilum of the lung are not enlarged; they contain a considerable amount of pigment.

The *heart* and *aorta* show no significant changes.

Diaphragm. There is no demonstrable communication between the abscess in the liver and the right pleural cavity.

Bacteriologic report. The culture from the content of the large intestine was negative for dysentery bacilli.

Microscopic examination: *Liver.* The wall of the abscess is composed of dense connective tissue in its deeper part where there are many widely distended capillaries. At the outer margin of the abscess wall are small accumulations of liver cells undergoing atrophy, and also many mononuclear wandering cells of considerable size, many containing yellow pigment, others having vacuolated cytoplasm. Bile ducts are also found; some of these have a wide lumen, in others the lumen is very small. In the more superficial part of the abscess the wall is less dense; it is infiltrated with small round cells and a few polymorphonuclear leucocytes. Upon the surface are large colonies of cocci. Throughout the entire abscess wall are many large cells, often with basophilic cytoplasm, and having oval or round, vesicular nuclei. In each of these nuclei is a very prominent deeply staining mass. These cells are to be seen both within the capillaries and in the connective tissue. Frequently they appear to be passing through the wall of the capillary or are lying against the endothelial lining of the vessel; in places also they seem to have just penetrated the capillary wall and are lying immediately adjacent to it on the outer side. Some of the endothelial cells lining the capillaries are swollen. The liver lobules immediately about the abscess are flattened, and the liver cells atrophic. There is some increase in connective tissue in the portal areas near the abscess, and groups of phagocytes containing hemosiderin are to be seen. At a distance from the abscess the liver parenchyma appears normal. An occasional small accumulation of polymorphonuclear leucocytes grouped around a necrotic cell is found in the sinusoids; these degenerating cells are apparently not in continuity with the liver cells or endothelium of the sinusoids but lie free within the lumen.

Intestine. The ulcers extend down to the submucosa. The base is composed of loose granulation tissue infiltrated with small mononuclear wandering cells. In the granulation tissue and within small blood vessels are large cells with vesicular nuclei, in each of which is to be seen a very striking intranuclear mass. The infiltration with wandering cells does not involve the underlying muscle. In one or two instances an arteriole lying in the superficial part of the ulcer has undergone necrosis and the hemorrhage probably came from such vessels. There is no regeneration of the epithelium at the margin of the ulcer.

Lung. The smaller bronchi are filled with an acute inflammatory exudate and the epithelium has disappeared in many instances from at least a portion of the wall. In other places the epithelium has become flattened and more squamous in type. The walls of these bronchi are infiltrated with wandering

cells of all varieties, and the infiltration extends into the adjacent alveolar septa which are wide. Many of these alveoli are filled with polymorphonuclear leucocytes. In some of the bronchi the exudate has undergone organization and the lumen is partly filled with a fibrous plug. The exudate in some of the alveoli also is undergoing organization. The capillaries are engorged.

In other areas the alveoli are filled with a recent acute inflammatory exudate. The septa of these alveoli are not infiltrated with wandering cells. In many alveoli the epithelium is distinctly cuboidal, and apparently attached to or continuous with the lining epithelial cells are very large cells with vesicular nuclei, each containing a prominent, deeply staining mass. Some of these cells lie free in the alveoli or in the exudate within them. Large cells, at times multinucleated, with intranuclear masses, are found within the smaller branches of the pulmonary artery, or lying just beneath the endothelium of these vessels. These cells are identical in appearance with the striking cells found in the liver and in the intestinal ulcers.

In the section from the right lower lobe the pleura is greatly increased in width, and here is found granulation tissue which is very dense. The capillaries in the granulation tissue are quite large; adjacent to the capillaries, and at times within them, are to be found many of the large cells with the prominent intranuclear masses. The alveoli in the immediate vicinity of this granulation tissue have thickened walls, they are lined with cuboidal epithelium, and their lumina are much smaller than normal.

In the section from the left lower lobe, the pleura is only slightly thickened, and there is a delicate fibrinous exudate upon it. In many parts of the section the alveoli are filled with polymorphonuclear leucocytes, and the epithelial cells lining these alveoli are larger than normal, but less cuboidal than in many parts of the section from the right lower lobe. These epithelial cells are deeply basophilic in their staining reactions. The alveolar septa are frequently thickened. In many places the alveolar space is filled with edema fluid, and in such alveoli the lining epithelium is quite prominent. These alveolar septa are not especially thickened. In yet other alveoli is compact fibrin with a few polymorphonuclear leucocytes. The smaller bronchi are frequently dilated, and their lumina are filled with polymorphonuclear leucocytes; the epithelium has undergone metaplasia. Throughout the section are many of the prominent cells with the large intranuclear bodies. These are found in the smaller branches of the pulmonary artery, in the capillaries or just beneath the endothelium of these vessels, and also free within the alveoli or in direct continuity with the epithelial cells lining the alveoli.

With the Gram stain, many gram-positive diplococci, often in pairs, are seen in the areas of acute inflammatory exudate.

Spleen. In the pulp are many polymorphonuclear leucocytes and hemorrhages. None of the large cells are found.

Adrenals. The cortical cells are depleted of lipoid. In one of the capillaries is a single large cell apparently undergoing degeneration.

Testes. There is no spermatogenesis. The membrana propia of some of the tubules is thickened and the interstitium is edematous. Small accumulations of lymphocytes, many of which are undergoing karyorrhexis, are found in the interstitial tissue.

Heart, kidneys, pancreas, and *prostate* are essentially normal.

Nuclear Inclusions

In the foregoing protocol, brief reference has been made to the intranuclear bodies present in intestine, liver and lungs. They were found, as has been noted, in large cells for the most part, isolated from the surrounding tissue, although in the lungs they appeared in some cases to be continuous with contiguous alveolar epithelial cells. Even with low magnification the inclusion-containing cells were conspicuous by virtue of their large size, measuring 25 micra. Usually they contained a single nucleus, but cells with two, three or four nuclei were seen. In these multinucleated cells inclusions were sometimes present in only one or more of the nuclei (Fig. 3).

The inclusions themselves varied in size, shape and, to a certain degree, in their staining reaction. The largest forms measured 11 micra in greatest dimension and completely filled the nuclear area with the exception of a narrow, clear zone sharply separating them from the deeply stained nuclear membrane. Projecting into this clear zone from the nuclear membrane were the deeply stained remnants of the chromatin material, often aggregated into a single lenticular clump. In addition smaller and less definitely stainable chromatin granules could be seen lining the nuclear membrane on its inner surface. The smaller forms, of which there were often several within the nucleus, were at times difficult to distinguish from the nucleoli and, as will be pointed out later, stained less distinctively.

The shape was most commonly spherical or ovoid, but elongated sausage-shaped forms were seen (Figs. 1 and 2). The structures appeared to have a certain plasticity, conforming to the shape of the nucleus. They generally were sharply outlined, but occasionally their margin was somewhat fuzzy and indistinct. Forms which were interpreted as degenerative showed fusion with the nuclear membrane, the latter also becoming indistinct.

The bodies showed little evidence of internal structure. At times the central portion stained more intensely; at times also one could see irregular areas of rarefaction. By none of the staining methods used, including Heidenhain's iron-hematoxylin, was it possible to identify minute granules within the inclusions.

The staining reactions conform closely to those given by Lipschütz [14] in his original article on herpetic inclusions. In addition to the reactions given by Lipschütz, it was found that the bodies stained

brilliantly red with Mallory's acid fuchsin — aniline blue, orange G, and that they did not retain the gentian violet in the Gram stain. With the Levaditi stain they became a uniform chestnut brown in contrast to the yellow of the remaining tissue. With Bensley's modification of Altman's mitochondrial stain the inclusions were vividly red. The cytoplasm of the inclusion-containing cells stained purplish with the hematoxylin-eosin, being in general more basophilic than the surrounding tissue cells. The cytoplasm appeared finely granular; but indistinct and well-defined granules were demonstrated only with the Mallory stain, and when present they were intensely fuchsinophilic. These cytoplasmic granules were not found in all of the inclusion-bearing cells. Stained with Scharlach R, some of the larger cells in the lung sections were found to contain finely divided fat globules within the cytoplasm; these did not extend completely to the surface but left a clear superficial zone. The outline of the cell was usually sharp, but it was sometimes possible to discern a paler staining fringe suggesting an ectosarc. One or two cells were found with plasmatic pseudopods, devoid of granules.

Location and Origin of the Cells

In the intestinal lesions the inclusion-containing cells are found in the stroma of the granulation tissue forming the base of the ulcers. Frequently they are near the blood vessels and indeed are often seen lying immediately beneath the endothelium, penetrating it, or within the lumen.

In the liver, they are most numerous in the granulation tissue lining the wall of the abscess, both intra- and extra-vascularly. A few degenerating forms were found in the hepatic sinuses at a distance from the lesions.

In the lung they are found projecting from the alveolar wall and interposed between unaffected alveolar epithelial cells; free in the alveolar cavities; and in the walls of the arteries and veins and within and without the lumina of the alveolar capillaries (Fig. 4). They are also found in the granulation tissue of the pleura. The large forms are not present in the bronchial epithelium or within the lumina of the bronchi.

The bodies as they have been described above, present little difficulty in their recognition. A closer study of the sections, especially those of the lung, discloses the presence of smaller intranuclear

inclusions within a large proportion of the alveolar and bronchial epithelial cells. These are distinguishable with difficulty from nucleoli. In sections stained with aniline fuchsin-methyl green, however, they retain the fuchsin whereas the nucleoli of the bronchial and alveolar epithelial cells, studied in twenty other cases, failed to give this reaction. One is therefore led to believe that these smaller forms may represent early stages in the formation of the larger inclusions.

As regards the origin of the cells it seems certain that there is more than one type of cell affected. In the lung the large inclusion-containing elements are frequently seen in direct continuity with neighboring alveolar epithelium. On the other hand the presence of cells with intranuclear bodies in the granulation tissue and within the blood vessels points to another source, obviously not epithelial. So alien in their appearance, however, are these cells that it is impossible to assign to them a definite origin from any one particular type of mesenchymal element. There is a striking tendency of those cells which are not of epithelial origin to localize in the vicinity of the blood vessels, or immediately beneath the endothelium, or even within the lumen. This suggests, but does not establish, their origin from the adventitial wandering cells.

Discussion

Since there is no possibility of carrying on an experimental study with the material from this case, an interpretation of the nature and significance of the inclusions must for the present rest upon the basis of similar observations recorded by others. There can be no doubt that the inclusions are identical in their morphology and staining reactions with the bodies seen by previous observers in the viscera of infants, and by Lipschütz and others in the tissues of spontaneous and experimental herpes, and in the various neural and visceral lesions produced by the herpetic and related viruses. Various possible interpretations thus present themselves:

1. The bodies (including those occurring in herpes and related conditions) represent merely a peculiar form of nuclear degeneration, not produced by a specific virus.
2. The bodies indicate the localization of the herpes virus or a similar virus within the nucleus of certain visceral cells.

It seems hardly necessary to consider seriously the possibility that the inclusions themselves or the inclusion-containing cells represent protozoan parasites. The mere fact that bodies of this appearance can be produced at will in a variety of tissues by the injection of herpetic virus, effectually disposes of this theory.

While it may be admitted that no positive proof has yet been brought that inclusions of this type are due to an intranuclear virus, there are several strong arguments against the first possibility, namely, that they are merely non-specific nuclear degenerations. They are present in cells which are evidently actively mobile. It seems unlikely that cells showing such marked nuclear degeneration should preserve their capacity for ameboid motion. Furthermore, individual cells are selectively involved whereas in the ordinary degeneration large numbers of the cellular elements are simultaneously affected. Again there are no obvious degenerative alterations in the cytoplasm of the inclusion-containing cells apart from the occasional presence of fat. On the basis of the above considerations, and above all because of the infrequency of their occurrence in routine pathological material, it seems safe to infer that the inclusions are not produced by a banal, non-specific nuclear degeneration.

There is but one argument, a very convincing one, however, in favor of the second possibility, that the inclusions in this case are caused by a virus identical with or closely related to the herpetic group. That argument is the morphological and tinctorial identity of the structures with those occurring in spontaneous and experimental herpetic lesions. We have recently had opportunity to compare the inclusions with those produced in the rabbit cornea and brain by the inoculation of the contents of a vesicle from a case of herpes labialis, occurring in one of the laboratory workers. The inclusions correspond in every particular.

Recent work has made it probable that viruses of this nature are more widely distributed, and perhaps more persistent, than had hitherto been suspected. Although it could not be established that the virus isolated by Rivers and Tillett [25] from cases of varicella was actually the cause of that disease, the virus obtained did produce in the skin, testicles and corneae of rabbits intranuclear inclusion-bodies apparently identical with those described in herpes, and situated for the most part within endothelial leucocytes as well as within the epithelial cells. Most interesting also is the transmissible virus

obtained by Miller, Andrewes and Swift [26] from the testes of rabbits inoculated with the blood and joint fluids of patients suffering from acute rheumatic fever, which had the power of producing inflammatory lesions with typical nuclear inclusions in the testes, skin, pericardium and heart muscle of inoculated animals. Still more interesting and puzzling is the isolation by Andrewes and Miller [27] of an apparently identical virus from the testes of supposedly normal rabbits. Flexner and Amoss [28] have also reported obtaining a very virulent neurotropic virus from the spinal fluid of a case of vascular and neural syphilis.

That man may harbor the herpes virus in a latent state is suggested by the experiments of Bastai and Busacca.[29] They examined the blood and spinal fluid of a large series of patients who had had no herpetic eruption for a long period. Positive results were obtained by corneal inoculation in rabbits in a large proportion of cases.

We are obviously at the beginning of our knowledge of this interesting group of diseases and it would be ill-advised and premature to say that the case presented illustrates a hitherto unrecognized disease caused by a virus akin to the herpes viruses. And yet, after studying the rather confused literature in this field, it seems less improbable than it did at first glance. There is first of all the closest possible morphological identity between the nuclear inclusions found in our case and those which are coming to be generally recognized as characteristic of the herpes-encephalitis type of virus. Secondly, there is growing evidence that the lesions produced by viruses of this type are not necessarily restricted to neuro-ectodermal structures, as maintained by Levaditi,[30] but may under suitable conditions be produced in such diverse tissues as trachea, bronchi, ovary, testis, adrenal (Goodpasture and Teague [31]). The virus may be present in the blood, spinal fluid or buccal secretions, and may be introduced into the body by direct inoculation of liver, pancreas, ovaries, thyroid, salivary glands, kidneys and spleen (Teissier, Gastinel and Reilly [32]). Even if we assume, as is probable, that the peculiar large cells with inclusions are to be interpreted as an infection with a virus of this group, it is still impossible to prove that this virus is the primary cause of the intestinal ulcers, liver abscess and pneumonia. The presence of bacteria in the liver abscess and pulmonary lesions makes it still more difficult to arrive at a conclusion. In favor of the view that the supposed virus is concerned in the lesions are:

First, the occurrence of the inclusion cells in numbers only at the site of the lesions; second, the fact that viruses of this type are capable of inciting an inflammatory reaction, in some cases of considerable severity; and third, that we are unable to attribute the changes to any other discoverable agent.

We are indebted to Dr. Walter W. Palmer and Dr. Allen O. Whipple for permission to transcribe the clinical record, and to Mr. Alfred Feinberg for the drawings.

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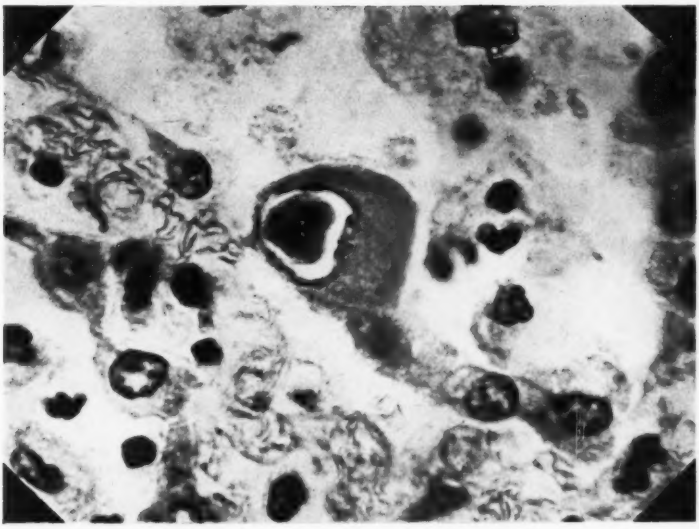
DESCRIPTION OF PLATES LXXII–LXXIII

Fig. 1. Cell with intranuclear inclusion, continuous with lining epithelium of lung alveolus. (Hematoxylin-eosin stain.) X 1050.

Fig. 2. Inclusion-containing cell lying within capillary in wall of liver abscess. (Hematoxylin-eosin stain.) X 1050.

Fig. 3. Lung: Multinucleated cell in alveolus. Two of nuclei contain typical inclusions. (Eosin-methylene blue stain.) Oc. 10x. Imm. 1/12.

Fig. 4. Lung: Two inclusion-containing cells lying just outside endothelium of a capillary. (Eosin-methylene blue stain.) Oc. 10x. Imm. 1/12.



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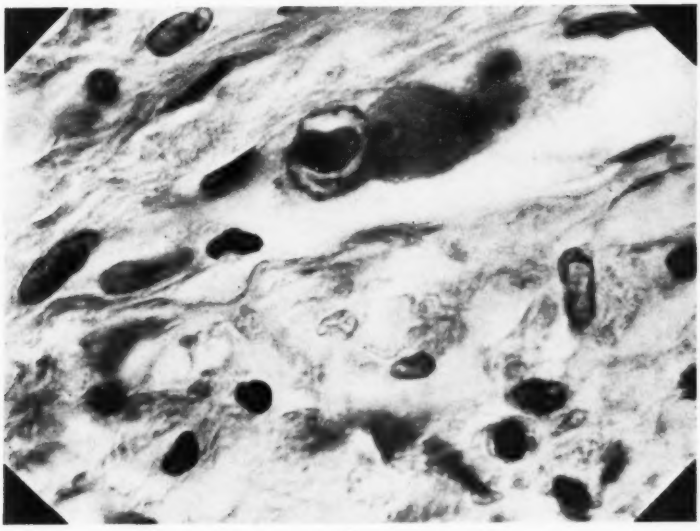

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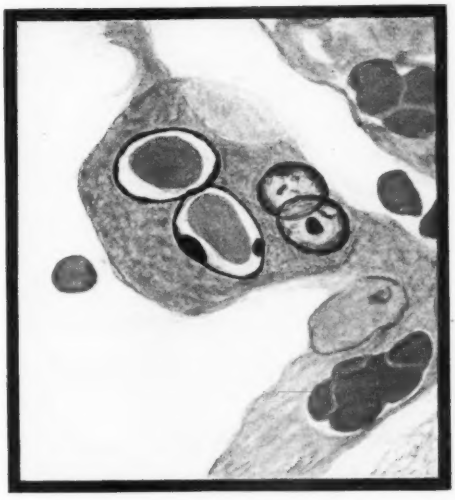

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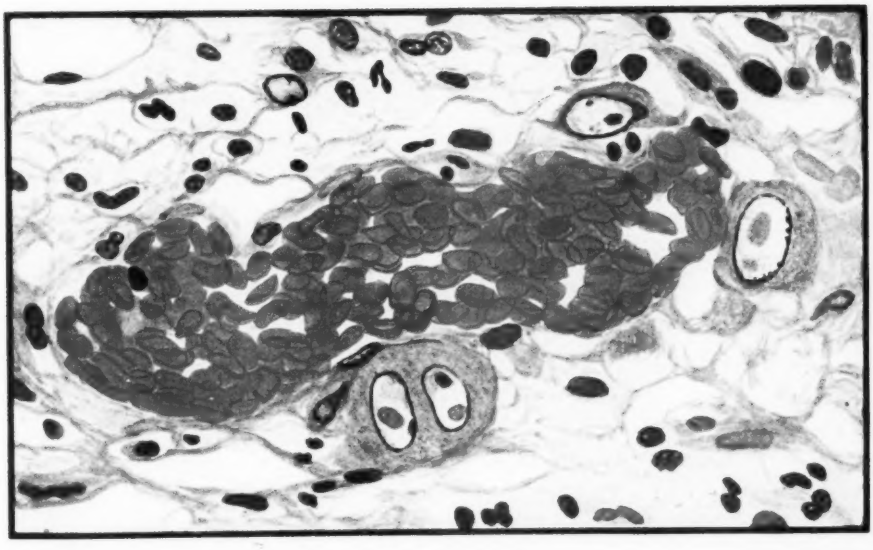

4

CONGENITAL HEART DISEASE WITH PARTIAL SITUS INVERSUS, ABSENCE OF INFERIOR VENA CAVA, AND OTHER ANOMALIES *

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Because of the unusual features of the case and the fact that a study of anomalies may help solve embryological phenomena, this report and discussion are submitted.

Patient, B.D.E., entered the hospital because of refusal to nurse, and cyanosis on crying.

Past History. The babe was full term and delivery spontaneous.

Birth weight, 7 lbs. Obstetrician noted no abnormalities. Breast fed.

Present Illness. Patient seemed well until after the first few weeks of life, when cyanosis was noted on crying. The attacks increased in frequency and degree. The patient gradually became more irritable and refused food. At the age of ten weeks rapid respirations were noted and the patient entered the hospital.

Physical Examination. Patient is a well developed, fairly well nourished baby girl ten weeks old. Cyanosis is marked in fingers and lips, slight on cheeks and forehead. Dyspnœic. Respiratory rate, 66 per minute. Cardiac dullness 1 cm. to the left of nipple line and 2.5 cm. to right of right sternal margin. Transverse cardiac dullness 8 cm. Signs of consolidation on lower right side of chest. Liver palpable on left side instead of right. Both heart sounds are altered and a murmur continues throughout cardiac cycle. Temperature 96°.

Clinical Course. Child refused to nurse, continued to be very irritable, and the cyanosis on exertion increased. Dyspnœa increased. Temperature rose to 96.6° on the day following admission and death occurred the second night.

Clinical Diagnosis. Bronchopneumonia, congenital heart disease.

NECROPSY REPORT

External Examination. Body is well developed and fairly well nourished, 54 cm. in length. There is a definite livid hue to entire body and less settling of blood posteriorly than usual. No edema. No discharge from any of the body cavities. Chest is rounded and full in upper part.

Peritoneal Cavity. About 200 c.c. (estimated) of clear, straw colored fluid. Right diaphragmatic dome is at the level of the 7th

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rib, and the left at the 7th interspace. Relations are as shown in Figs. 1 and 2, and will be described in detail below.

Chest Cavities. Pleural cavities are moist but there is no excess fluid. Parietal pleura is normal. Pericardial sac appears normal. It contains about 6 c.c. (estimated) of clear, straw colored fluid.

Mediastinum. Thymic tissue is recognized and appears normal in amount and consistence. Lymph nodes are slightly large and red. Relations described below.

The viscera are preserved *en bloc* for further dissection and study, and, therefore, not weighed.

Heart and Great Vessels. The position of the heart is normal. The great vessels are anomalous, as is shown in Figs. 3, 4 and 5. The

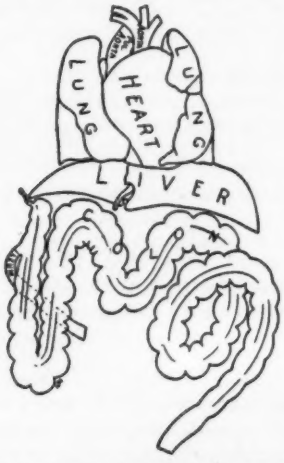

Fig. 1. Sketch at autopsy showing the redundant colon; the ileum passing posterior to the colon to enter it from the lateral side; the cecum and appendix in the right hypochondrium and the large left lobe of the liver.

common iliac and renal veins pass posterior to the aorta and empty into the azygos vein, which is normally placed but comparable to the inferior vena cava in size (Fig. 3). The inferior vena cava is absent save for the short vessel passing from the liver to the right atrium.

The systemic and pulmonic aortae are transposed and both arise from the left ventricle (Fig. 4). The pulmonary vessel is about twice the size of the systemic but the walls are about equal in thick-

ness. The right subclavian and right and left common carotid arteries are branches of a common stem. The left innominate vein does not join the right but passes downward, joins the coronary sinus, which is large, and empties into the right atrium (Figs. 4, 5 and 7). The ductus arteriosus (Fig. 5) is still functioning and measures 6 mm. in diameter.

The right atrium is dilated and hypertrophied, its wall being 1 mm. to 4 mm. in thickness. It receives blood from the superior and inferior venae cavae and the coronary sinus. Blood may leave this chamber through the patent foramen ovale, which is 13 mm. in diameter, through a large opening into the left ventricle and through a slit-like opening 1 x 3 mm. into the right ventricle (Figs. 6, 7 and 8). The atrial septum is not applied to the base of the ventricles but has a free margin. The annulus ovalis is absent in the anterior superior portion of the septum.

The right ventricle is small and under-developed, the outer wall being 2 mm. to 4 mm. in thickness. There are no papillary muscles in the right ventricle and its valve is incorporated with the mitral. The only entrance is the above mentioned slit. The only outlet is through the space where the pars membranacea septi failed to form.

The left ventricle is dilated and hypertrophied, its walls being 8 mm. to 12 mm. in thickness, and the moderator bands are very large. The mitral and tricuspid valves are fused into a large funnel-shaped curtain attached at the base of the ventricles and emptying into the left ventricle (Figs. 7 and 8). There are three partially formed leaflets, a right and left anterior and a posterior one. A row of papillary muscles is attached by long chordae tendineae to the left leaflet and a part of the posterior one. The right leaflet has its base attached to the outer wall of the right ventricle, covers the right chamber completely save for the slit-like opening, lies across the free margin of the ventricular septum and is attached on the left side of the ventricular septum by short chordae tendineae and miniature papillary muscles. The left ventricle has two outlets, the systemic and pulmonic aortae, the latter being to the right of the former (Fig. 9).

One coronary artery arises from the left posterior sinus of the aorta. The right and left coronary arteries are branches of this one vessel and the left gives off the circumflex branch. The final distribution is similar to the normal (Fig. 5).

The direction of muscle fibres is similar to the normal.

Lungs. The lungs are atypically lobed and fissured. They are dark, mottled red in color, firm, not crepitant save for a small part of the right upper lobe and the anterior part of the left upper lobe.

Liver. The liver, gallbladder, stomach, duodenum, pancreas and spleen are transposed so as to form an optical image of the normal, as is shown in Fig. 2. The left lobe of the liver is large; the hilum, gallbladder and spigelian lobe are to the left of the umbilical cleft. The inferior vena cava extends from the right lobe of the liver to the right atrium. The common duct empties into the second portion of the duodenum anteriorly.

Pancreas. The head is in the arch of the duodenum and the tail extends to the left. The ducts of the ventral and dorsal portions both persist, the latter emptying caudad to the ampulla of Vater instead of cephalad, as usual. The ventral duct joins the bile duct at its ampulla.

Spleen. There are six separate masses of splenic tissue (Fig. 3). Four are small and spherical, measuring 2 mm., 2 mm., 4 mm., and 10 mm. respectively in diameter. The remaining two are similar to the normal in shape, measuring 3.5 cm. x 2 cm. x 1 cm. and 4.5 cm. x 2.5 cm. x 1 cm. respectively. The hila face a common center. The largest one has a deep transverse fissure.

Gastro-intestinal tract. The esophagus is slightly to the right of the trachea, instead of to the left, which is in keeping with the transposed position of the stomach (Figs. 2, 3 and 4). The duodenum arches to the left so that the second portion is to the left of the second lumbar vertebra. The common duct enters the second portion anteriorly and is joined at the ampulla by the duct of the ventral pancreas (Wirsung's duct). The accessory pancreatic duct (duct of Santorini), or the dorsal pancreatic duct, enters medially and caudad to the ampulla of Vater, forming an ampulla in the mucosa.

The small intestine lies coiled in the lower and right part of the abdomen. The root of the mesentery extends downward and to the left, but is almost horizontally placed.

The colon is redundant. The cecum and appendix are in the right hypochondrium, extend upward and are in relation to the right small lobe of the liver. From here the colon passes downward, then left and upward, forming a "U" loop. The ileum passes upward, under both limbs of the "U" loop in the first portion of the colon, to enter

it laterally, as shown in Fig. 1. The transverse colon is long and sags. The portion of the colon in the left hypochondrium forms a complete spiral turn and then passes downward in the usual way.

The kidneys, ureters and pelvic viscera are not remarkable.

Pathologic diagnoses. Bronchopneumonia, bilateral, confluent; malformations of heart, great vessels and colon; situs inversus of liver, gallbladder, stomach, duodenum, esophagus, pancreas and spleen; persistent dorsal pancreatic duct.

Discussion

Normally, the inferior vena cava is a composite vessel, being formed from below upward by the union of the following vessels: the lower right veins of the prevertebral plexus (formerly this portion was thought to be a part of the posterior cardinal); the lower part of the right subcardinal; vein of the plica vena cava (caval mesentery) and the primitive right vitelline. The kidney develops opposite the anastamosis of the subcardinals in tissue containing a venous plexus which drains into them. A second venous plexus is found posterior to the above mentioned one and is called the prevertebral plexus. If the kidney develops around the latter plexus instead of the former it is possible for its vein to drain into the posterior vessels which form the azygos [1] and it will then be the exact condition described above. The subcardinal as well as the posterior cardinal veins have atrophied. It is also possible that the kidney developed in its normal position and the atrophy of the subcardinals occurred first, forcing the kidney veins to drain into the prevertebral plexus.

The position of the aortae is such that the systemic vessel is entirely on the left side. This may suggest that the first portion of the arch was derived from the left ventral aorta and the fusion and atrophy of the ventral aortic septae and possibly the rotation failed to occur. It is, however, not possible to trace either of the ventral aortae to the adult heart. In order to bring the aortae in proper relation in the above case, there must be a dextral tortion of 270°. It is interesting in this connection to remember Rokitansky's [2] sixteen possible forms of transposition, some of which were found after he had described them. Another possible explanation of aortic transposition has its foundation in the possibility that the first and second portions of

the adult aorta are normally a composite vessel and that the twined structures do not depend entirely on tortion for their relations.

The common stem for the three arteries, right subclavian and both common carotids, is a very common anomaly in man, occurring once in ten according to Parsons,[3] and is normal for rodents and other animals. The normal condition for the llama and giraffe is similar, differing only in this, that the common carotids have a long bicarotid stem. Morever, it is possible, as suggested by Lewis,[4] that the arrangement is a sign of Nature's effort to maintain symmetry which is so nearly, but not entirely, attained in the sheep. The arrangement is properly ascribed to migration of the left common carotid. The reason for migration is obscure, but the fact of migration is clearly shown by Jackson.[5]

The course of the left innominate vein is plainly the persistence of an embryonic structure, the left anterior cardinal vein. The anastomosing branch between the anterior cardinals, which develops to carry the blood from the left into the right, was not found. Its failure to form or to develop, as the case may be, would seem to explain the persistence of the distal portion of the left anterior cardinal and the course of its blood through the left duct of Cuvier (left common cardinal vein continued). The absent anastomosis may be result and not cause.

The ductus arteriosus is also a persistent embryonic structure — the distal part of the left sixth aortic arch. Whether or not the narrowing of the aorta in the second portion of the arch proximal to the opening of the ductus is an effect or a cause of its persistence is not demonstrated. In case of atresia of either aorta, the function of the persistent duct might seem to be clear. But in case of constriction of the aorta distal to the duct, its persistence on the ground of furnishing necessary blood is not explained. Such a condition is recorded at Iowa City.[6] In a series of 142 cases of aortic constriction mentioned by Abbott,[7] the ductus arteriosus was patent in only thirteen.

The lack of development of the right ventricle seems to be the result of a lack of opportunity for work. The only inlet is small and insufficient to carry blood to the ventricle, while its only outlet, the unclosed pars membranacea septi, would be closed during systole by pressure of the valve leaflet on the free edge of the septum. The enormous left ventricle might be thought of as a result of overwork, since the right ventricle is practically cut out of the circulation and

the heart functions as a bi-atrial-triloculate heart. The large septal defects allowing free passage of blood from one atrium to the other would seem to equalize the work of them. In this connection it is interesting to note the cases cited by Abbott,[7] as recorded by Simmonds,[8] Effron,[9] Kalb [10] and Ratner,[11] in which there was cardiac congenital hypertrophy without apparent cause. Hypertrophy without demonstrable cause is also found in adults, but is frequently spoken of as "so-called idiopathic hypertrophy." Certainly all of the influences which bring about hypertrophy are not clear.

The position of the atrial septum is normal. The interatrial foramen (Foramen primum) resulted from a failure of fusion with the base of the ventricles. The septum is smooth on either side and may represent the septum primum with an arrest of development before the formation of the posterior portion and a failure of formation of the septum secundum. The foramen ovale is not yet completed, since the antero-superior part of the annulus is absent. The opening here, then, is the foramen secundum, and should be called "foramen ovale" when the annulus is complete.

The ventricular partition is incomplete in that the pars membranacea septi failed to form and the muscular portion is placed to the right of its usual position. If the ventricular septum had grown from the tubercle between the aortae (Fig. 9), the functioning, assuming the presence of atrio-ventricular valves, would have been perfect and the condition would conform to the type, "corrected transposition," as described by Rokitansky.[2]

The formation of one valve at the base of the ventricles was probably influenced by the malposition of the ventricular septum.

The transposition of a part of the viscera is interesting in that it indicates that the forces causing rotation are probably different for the different viscera. The dextral rotation is in keeping with the common direction of the spiral in gastropod mollusks, yet a few of these turn sinistrally. However, the forces at work in these lower forms are also obscure. It may be suggested that in the above case the position of the esophagus is secondary to that of the stomach. The stomach may depend on the direction of the arch of the duodenum, which in turn may be dependent on the development of the liver and gallbladder. The hilum, being to the left of the umbilical fissure, is in keeping with the notion that the persisting vessels on the left instead of on the right have influenced the left part of the

liver to develop larger. The efferent vessel (vena cava), however, leaves the right lobe as normally.

The accessory pancreatic duct is the persistence of the dorsal pancreatic duct, as described by Santorini. It departs from the normal in that it has developed caudad to the ventral pancreas instead of cephalad, as usual. The placement of the ampulla of Vater and the duct of Wirsung (ventral pancreatic duct) cannot be explained on a simple basis of sinistral rotation.

Summary

The case presented may be summarized as: congenital heart disease comprising a patent foramen primum and secundum, patent pars membranacea septi, one atrio-ventricular valve emptying into the left ventricle, misplaced ventricular septum, both aortae arising from the left ventricle, hypertrophy of the left ventricle and right atrium; anomalous vessels comprising absence of inferior vena cava with the renal and common iliac veins emptying into the azygos vein, absence of the transverse portion of the left innominate vein which empties into the coronary sinus, a common stem for the right subclavian and both common carotid arteries, transposition of the aortae, persistent ductus arteriosus, and both coronary arteries arising from a common stem; *situs inversus* of liver, gallbladder, hilum of liver, esophagus, stomach, duodenum, pancreas and spleen; accessory pancreatic duct and spleens.

Explanation is offered as involving the following processes:

1. Persistence of embryonic structures, namely, interventricular foramen, foramen primum (interatrial foramen), foramen secundum (later called the foramen ovale), left anterior cardinal vein, dorsal portion of left sixth aortic arch (ductus arteriosus), dorsal pancreatic duct (duct of Santorini), branches of prevertebral plexus forming the renal veins.

2. Failure of development, namely, anastomosis of the anterior cardinal veins forming the left innominate vein; the lower right veins of the prevertebral plexus, right subcardinal, vein of plica vena cava, and primitive right vitelline vein forming the lower part of the inferior vena cava; one of the atrio-ventricular valves; septum secundum; posterior part of septum primum; pars membranacea septi.

3. Misplacement, namely, left common carotid artery, ventricular septum, ileum, lower part of ascending colon, cecum and appendix.

4. Reversed rotation, namely, esophagus, stomach, duodenum, pancreas and spleen.

5. Development of accessory structures, namely, multiple spleens.

6. Hypertrophy of azygos vein, left ventricle and right atrium.

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DESCRIPTION OF PLATES LXXIV — LXXVII

Fig. 2. Drawing of the ventral view of the transposed abdominal viscera. They form an optical image of the normal. The left lobe of the liver is large. The gallbladder, hilum and spigelian lobe are to the left of the umbilical cleft. Both pancreatic ducts persist. Sp., spleens; St., stomach; Li., liver; S. L., spigelian lobe; P., pancreas; D.V.P., duct of ventral pancreas (Wirsung's); D.D.P., duct of dorsal pancreas (Santorini's); D., duodenum; P. V., portal vein.

Fig. 3. Posterior view of viscera of the trunk. The common iliac veins and renal veins empty into the azygos vein. The esophagus is slightly to the right. L.C.I.V., left common iliac vein; R.C.I.V., right common iliac vein; L.U., left ureter; R.U., right ureter; L.R.V., left renal vein; R.R.V., right renal vein; A., aorta; A.V., azygos vein; R.I.V., right innominate vein; E., esophagus; L.S.A., left subclavian artery; L.C.C.A., left common carotid artery; L.I.V., left innominate vein; D.A., ductus arteriosus.

Fig. 4. Ventral view of chest viscera with right lung retracted. The right atrium and left ventricle are hypertrophied and dilated, the right ventricle rudimentary. The aortae are transposed. R.A., right atrium; R.V., right ventricle; I.V.G., interventricular groove; L.V., left ventricle; L.I.V., left innominate vein; L.S.A., left subclavian artery; L.C.C.A., left common carotid artery; T., trachea; E., esophagus; R.C.C.A., right common cartid artery; R.S.A., right subclavian artery; R.I.V., right innominate vein (superior vena cava.)

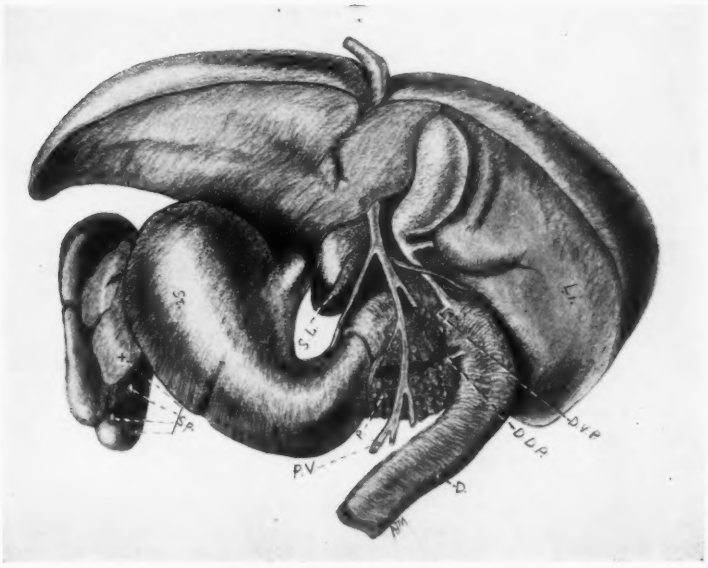
Fig. 5. Left view of heart and great vessels with a window in left atrium which is rudimentary. The two orifices in the atrial septum have persisted. The left innominate vein still joins the coronary sinus. The ductus arteriosus persists. Three large arteries arise from the aorta by a common stem. The coronary arteries arise from a common stem. C.S., coronary sinus; I.V., inferior vena cava; L.P.V., left pulmonary vein; P.A., pulmonary artery; D.A., ductus arteriosus; L.S.A., left subclavian artery; L.I.V., left innominate vein; A., aorta; L.C.C.A., left common carotid artery; R.C.C.A., right common carotid artery; R.S.A., right subclavian artery; F.O., foramen ovale; F.P., foramen primum; C.A., coronary artery.

Fig. 6. Right view of heart and great vessels with window in right atrium showing atrial septum. I.V., inferior vena cava; S.V., sinus venosus orifice; F.P., foramen primum; R.V., right ventricle; L.V., left ventricle; F. O., foramen ovale (foramen secundum); R.A., right auricle; R.C.C.A., right common carotid artery; R.S.A., right subclavian artery; L.C.C.A., left common carotid artery; R.I.V., right innominate vein (superior vena cava); A.V., azygos vein; P.A., pulmonary artery; P.V., pulmonary vein; P.S., indicates plane of section of heart for Figs. 7, 8 and 9.

Fig. 7. Anterior view of posterior portion of heart sectioned as shown in Fig. 6. The under developed and over developed portions of the heart are shown. The atrio-ventricular valve is funnel-shaped and empties into the left side. A slit-like opening allows some blood to go into the right ventricle, which has no outlet save through the patent pars membranacea. The chordae tendineae attachments are shown here and in Fig. 8. L.V., left ventricle; L.A., left atrium; C.S., coronary sinus; P.V., pulmonary vein; A.S., atrial septum; R.A., right atrium; I.V.C., inferior vena cava; S.V., sinus venosus; S., slit-like opening in atrio-ventricular valve opening into right ventricle; V. S., ventricular septum; R.V., right ventricle.

Fig. 8. Posterior view of anterior portion of heart. V.S., ventricular septum; R.V., right ventricle; A.V.V., atrio-ventricular valve; F.O., foramen ovale; A.S., atrial septum; A.L., auricular lumen; R.A., right auricle; R.I.V., right innominate vein (superior vena cava); P.V., pulmonary vein; P.A., pulmonary aorta; A., aorta; L. A., left auricle; C.S., coronary sinus.

Fig. 9. Same as Fig. 8 with the valve flap dissected off to show the orifices of the aortae and the free edge of the ventricular septum, which is not in the same plane as the atrial septum, being misplaced to the right. V.S., ventricular septum; A.V.V., atrio-ventricular valve; P.A., pulmonary aorta; A., aorta; O.P.A., orifice of pulmonary aorta; E.C., endocardial cushion; O.A., orifice of systemic aorta.



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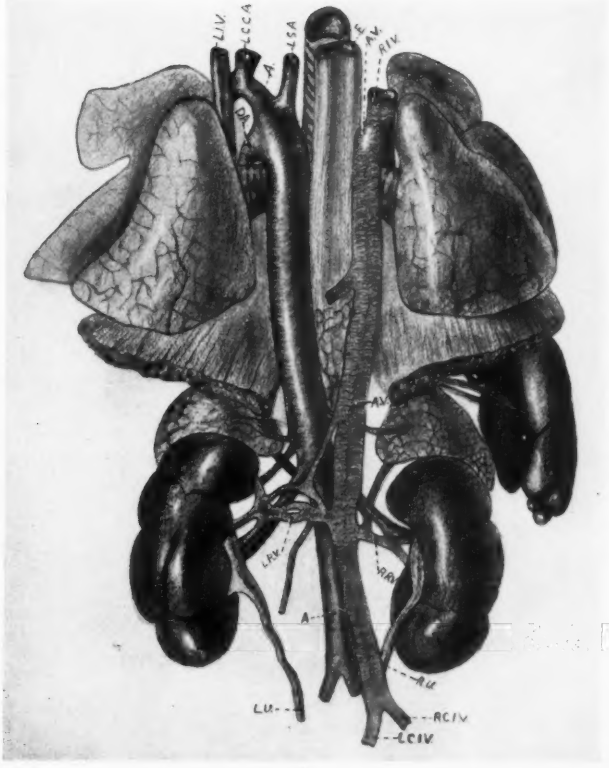

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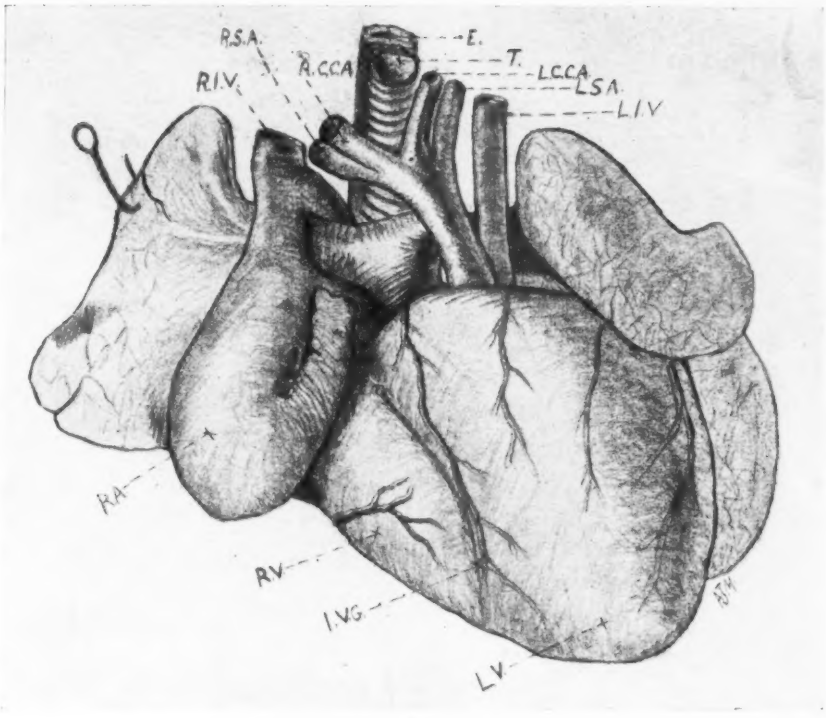
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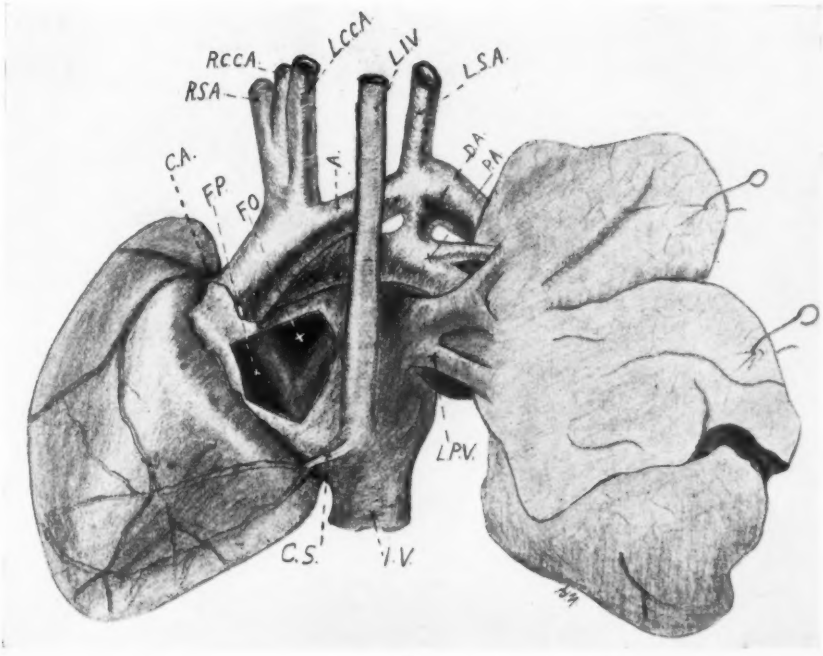
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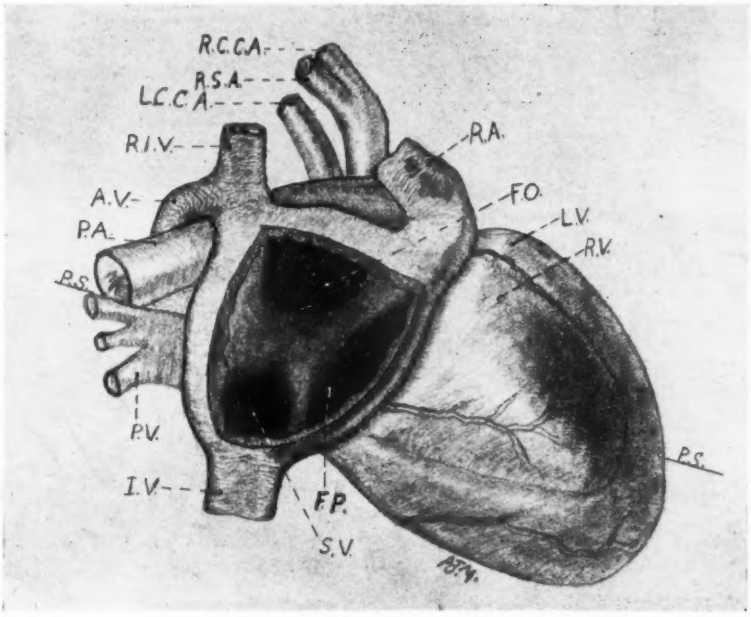
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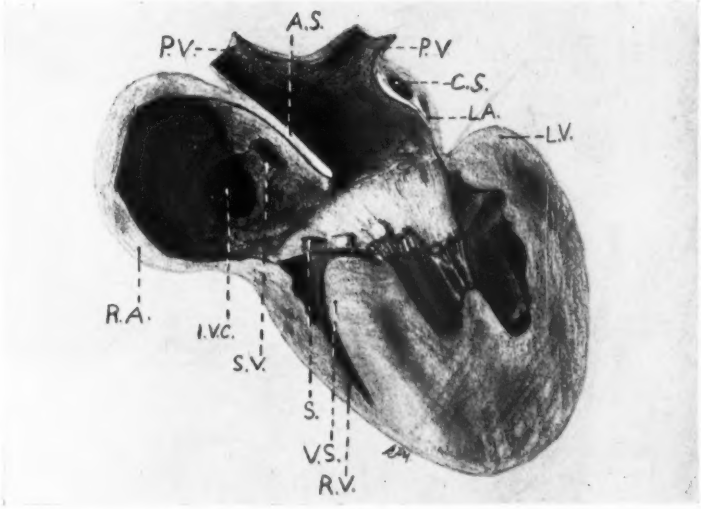
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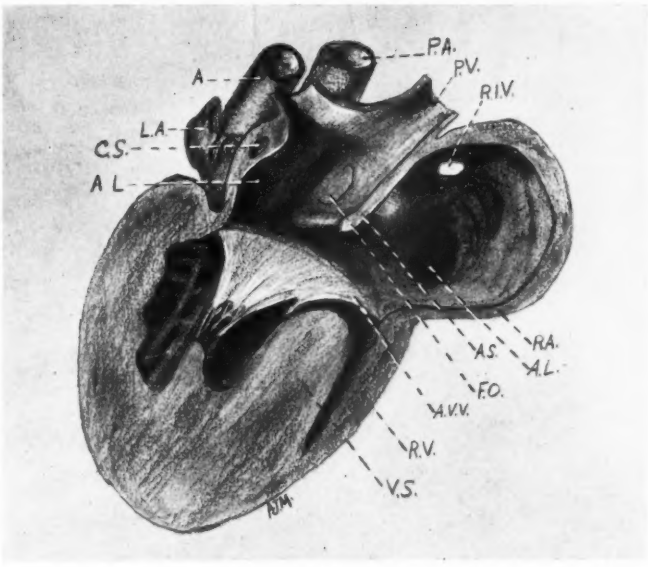

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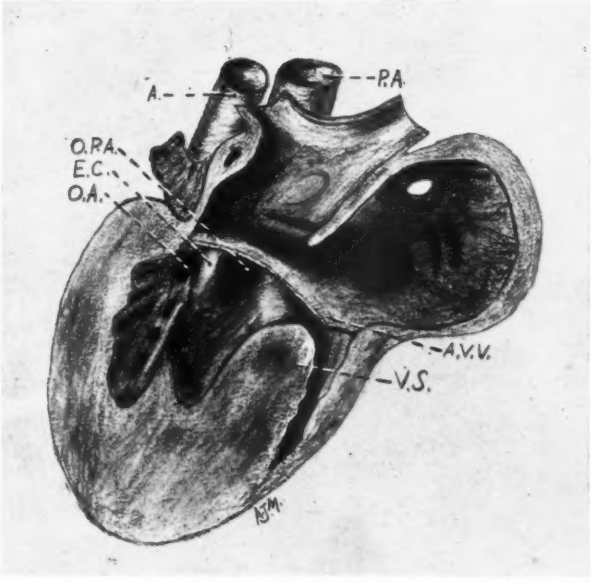

9

PROGRESSIVE ALCOHOLIC CIRRHOSIS *

REPORT OF FOUR CASES

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Since the time of Vesalius, who was the first to describe cirrhosis, the literature on the subject has been accumulating. Although volumes have been written, most accounts deal with the end stages of the disease. This seems the more remarkable when one considers that a study of the earlier stages gives greater promise of revealing the true nature of the process than does a study of the late stages. As Mallory [1] says, "In some organs the late stages of certain lesions have received more attention than the beginning of these lesions. The emphasis has been placed on the wrong end of the process. The statement is particularly true of the inflammatory changes in the liver and in the central nervous system." It is with these facts in mind that the present study is presented.

Diligent search of the literature has brought to light only a very few papers which deal specifically with the progressive stages of cirrhosis. Hawkins [2] regards the process as essentially inflammatory. He says, "If specimens of early cirrhosis are examined from cases in which death has occurred from other causes, no doubt can be entertained that the interstitial change is essentially an inflammatory one, and that it has its starting point around the branches of the portal vein at a time when the appearance of the degeneration of the hepatic tissue is either scanty or absent." Hawkins evidently underestimates the importance of the degenerative processes in the liver cells.

Mallory [1] in his study of the early lesions has paid particular attention to the peculiar form of necrosis of the liver cells which occurs in alcoholic cirrhosis and which seems to be characteristic of it. "The cytoplasm of the cell first undergoes a degenerative change in consequence of which an irregular, coarse, hyaline meshwork appears in it. . . . These processes (hyaline degeneration ending in necrosis, leucocytic infiltration, regeneration of liver cells, increase in the amount of connective tissue) when extensive, diffuse, and acute,

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lead to considerable increase in the size and weight of the liver." Mallory [3] states further, in his textbook, that there is a primary injury to the liver cells which is followed by more or less regeneration of them. In this he is in accord with those who have emphasized the importance of the regenerative processes in the disease. He goes on to say, "On the other hand, we have in alcoholic cirrhosis, around and invading each necrotic cell, an acute, inflammatory exudate of leucocytes which must cause more or less stretching of the connective tissue." This injury, he believes, results in the proliferation of fibroblasts which is mechanical, rather than toxic in origin.

Stengel and Kern [4] are doubtful as to the relative importance of the degenerative and inflammatory processes in the early lesions of cirrhosis. They say, "Fibrous tissue hyperplasia follows the injury to the parenchymal cells and may be simply a replacement fibrosis. In some cases, however, the evidences of inflammatory change (interlobular round cell infiltration, congestion and beginning fibrosis) are out of proportion to the evidences of cellular degeneration."

From these brief references it seems evident that there is still considerable difference of opinion in regard to the exact mode of development of the histologic changes in the liver in this disease.

Source of Material

In the present study four cases of cirrhosis showing relatively early progressive lesions in the liver have been utilized. These were selected from among about 150 cases of cirrhosis in over 3000 autopsies performed in the Department of Pathology of the Stanford University Medical School. The case histories and autopsy findings have been summarized and a careful histologic study of the liver made in each case.

Case 1

XX. 43. M.S., a white washerwoman, age 39 years, came to the San Francisco Hospital complaining of being unable to do sufficient work to support herself.

She had been married for eight years but later had separated from her husband. Two children had died of "spasms," one at one year, the other at 14 months. One child, 14 years old, is living. According to the patient's statement she had had no miscarriages. She denied ever having had any venereal infection.

She had never had any symptoms suggestive of disease of the liver. About one month before her entrance the patient began to cough and raise a small amount of sputum. She had lost weight since the onset of the cough, had felt weak and had tired very easily. Her voice had been husky for the past two

years but had not grown worse. The woman stated that she had used alcohol quite steadily for several years, wine with meals and one or two drinks of whiskey every day.

On clinical examination she proved to be a well-developed but undernourished woman of about 40 years. Her pupils were irregular and did not react to light or accommodation. Examination of the chest revealed the usual signs of pulmonary tuberculosis.

The abdomen was symmetrical. The liver dullness extended from the fifth rib to 12 cm. below the costal margin in the right mid-clavicular line. The lower border was notched and felt like liver edge; the surface was rough but not distinctly nodular. The liver was tender. The knee jerks were somewhat exaggerated.

The blood picture was normal except for a slight leucocytosis. The hemoglobin content was 95 per cent. The red blood cells numbered 4,500,000 per c.mm. and the white blood corpuscles 14,000 with a normal differential count. The urine contained a cloud of albumin and many pus cells. No red blood cells or casts were found in the sediment. Examination of the sputum revealed the presence of tubercle bacilli. The Wassermann reaction of the blood was triple positive.

Clinical diagnoses. Pulmonary tuberculosis, syphilis, cirrhosis of the liver.

Anatomic diagnoses. Syphilis of the aorta; aneurism of the arch perforated into the left bronchus; hemorrhage into the lungs; syphilis of the spinal cord; tuberculosis of the lungs, chronic; bronchiectasis; alcoholism, chronic; cirrhosis of the liver; carcinoma of the uterus (cervix); peritonitis, pelvic; suicide with a cutting instrument, attempted.

Abstract of autopsy record. The body is that of a strongly built, rather poorly nourished woman. The liver projects four fingerbreadths below the free margin of the ribs in the right mammillary line and two in the left. No fluid is present in the peritoneal cavity. The heart is less than one-half normal size; the heart muscle is thin and flabby. The wall of the aorta at its base is very much thickened, measuring about 3 mm. in thickness. The intima is full of small yellow spots and contains some scars.

The bronchi are filled with blood. Scarring and old caseous areas are found in the region of both apices with some recent tubercles in other parts of the lungs. The tonsils are small; the left one contains a small cavity filled with thick, viscid pus.

The arch of the aorta shows very marked thickening and extensive cicatrization. On the lower surface of the arch is an aneurismic sac about 7 cm. in diameter. The aneurism has perforated into the left bronchus.

The spleen is normal.

The cervix uteri shows a deep ulceration with indurated base.

The stomach contains some clotted blood. The gallbladder and biliary passages are normal.

The *liver* is rather large, measuring 26 x 26 x 7 cm. The surface is slightly granular. The liver tissue is quite firm, cutting with some difficulty. The cut surface shows a slight thickening of the periportal connective tissue.

Histologic examination. Microscopic examination of the cord reveals an early syphilitic inflammation of the meninges. Similarly sections of the aorta show the usual lesions of a syphilitic aortitis.

Liver. The capsule is of normal thickness. Within the liver there is a moderate increase in the periportal connective tissue. The new fibrous tissue has a patchy distribution and is arranged in roughly triangular or star-shaped areas from which finer fibers radiate out into the parenchyma enclosing small groups of liver cells (Fig. 1). The enmeshed liver cells show evidences of degeneration. Many of these cells contain hyaline masses in their cytoplasm; in others the cytoplasm is coarse, granular and vacuolate. The nuclei are irregular in shape and often swollen. They stain unevenly or in many cases they are absent altogether. Although the cell destruction is not so pronounced as in some of our other cases nearly every fibrous area contains groups of disintegrating cells. The development of collagen fibrils in the various areas of fibrosis is moderately advanced but the fibroblastic cells are still large and numerous.

There is evidence on every hand of widespread hyperplasia of the liver cells. Many areas are seen in which the cells have every mark of having been recently formed; they are well-filled, turgid cells; the cytoplasm is finely granular; the nuclei are of uniform size and staining. Many other cell-groups are found in which the cells appear to be recently formed but have a very pale cytoplasm which seems to be indefinitely vacuolate. These are, probably, cells loaded with glycogen. There is also a well-marked, rather diffuse, fatty infiltration of the liver cells as can be seen in Fig. 1.

Practically all of the new fibrous tissue is heavily infiltrated with polymorphonuclear leucocytes and lymphocytes. In many places the infiltration is quite marked and, as nearly as one can estimate, there are approximately equal numbers of neutrophilic polymorphonuclear leucocytes and lymphocytes.

Most interesting are the foci where the lesions are evidently quite recent (Fig. 2). Here many of the liver cells show hyaline degeneration and beginning necrosis. The spaces between them are being invaded by proliferating fibroblasts and there is a more or less heavy infiltration of the diseased tissue with polymorphonuclear leucocytes. The changes in the connective tissue are quite evidently of an inflammatory nature.

The "bile ducts" show only slight evidence of proliferation in or near the fibrous areas in this liver. Although there is evidence of widespread regeneration of the liver cells there is little to indicate that the "bile ducts" have played any appreciable rôle in the process.

The whole picture of this liver is one of considerable cell destruction and of extensive regeneration of the liver cells with a recent well-marked progressive inflammatory fibrosis. Some fatty infiltration is also present.

Bacteria were searched for in sections of liver stained with the Giemsa stain but none was found.

Case 2

XVII. 37. O. G. was an American teamster, 38 years old, single. He came to the San Francisco Hospital complaining of pains in the abdomen and swelling of the legs.

He had had "malaria" three years before and had been troubled with headaches for the past six months. His present illness began one month ago with pains in the abdomen and swelling of the legs. When the patient arrived at the hospital he was in a stuporous condition and his mentality remained clouded until his death. He had been a heavy drinker and he had been drinking to excess immediately prior to his entrance to the hospital.

Clinical examination showed jaundice. The liver edge was palpable and evidences of collateral circulation could be seen in the anterior abdominal wall. Ascites was marked and eight liters of clear fluid were removed by paracentesis. The calves of his legs were tender. The knee jerks and ankle jerks were absent on both sides.

The patient gradually grew worse and died as a result of his severe alcoholic intoxication.

His blood showed a slight secondary anemia and, toward the end, a high leucocytosis. The hemoglobin content was 80 per cent. The red cells numbered 4,000,000 in a c.mm. The Wassermann test of the blood and spinal fluid was negative.

Clinical diagnoses. Carcinoma of the liver; chronic alcoholism; alcoholic polyneuritis; Korsakow's psychosis; wrist and foot drop.

Anatomic diagnoses. Alcoholism; alcoholic psychosis; alcoholic neuritis; atrophy optic nerve; gastritis, chronic; cirrhosis of the liver (subacute); varices in the lower esophagus; pancreatitis with fat necroses; ulcers of the leg, healed; exophthalmus; tuberculosis of lungs, chronic, healed.

Abstract of autopsy record. The body is that of a fairly strongly built, extremely emaciated man. There is marked jaundice of the entire body. The left optic nerve shows a nearly complete gray atrophy. The abdomen is much distended laterally and contains about three liters of clear, greenish-yellow fluid with a few flakes of fibrin. The peritoneum is slightly hyperemic and moist. The liver extends slightly below its normal position. The heart is about three-quarters normal size.

There are old adhesions about the spleen which is slightly enlarged, measuring 12.5 x 10 x 4.5 cm. Its cut surface shows passive congestion and normal markings. There are some adhesions at the tail of the pancreas and the peritoneum over both kidneys is thickened. The lymph nodes at the hilum of the liver are slightly enlarged and bile-stained. The gallbladder contains thick, viscid, clear bile. The bile ducts are normal and patent.

The capsule of the *liver* is fairly normal except on the lower surface of the

left lobe where there is slight diffuse thickening with more marked fibrosis about the lymph vessels. The liver is somewhat above normal size, measuring 27 x 22 x 10 cm. The surface is smooth and of a greenish-gray color in which there are a large number of small, bright, yellow spots 1–2 mm. in diameter. The liver cuts with great difficulty. Its cut surface is smooth and shows numerous yellow spots with narrow gray bands between them. The liver substance is very heavy.

The pancreas is somewhat indurated and full of small fat necroses, the largest about 3 mm. in diameter.

Histologic examination.

Liver. The capsule is regular and of normal thickness, while the liver parenchyma presents a variegated picture. The lobules are greatly broken, in many places almost destroyed. Replacing the liver cells or ramifying among them are strands of recent, very cellular connective tissue with a marked cellular infiltration consisting largely of polymorphonuclear leucocytes (Fig. 4). This connective tissue is little beyond the fibroblast stage. The new connective tissue appears quite edematous. The periportal connective tissue is increased considerably in amount. It also is very cellular, yet it is somewhat older than the connective tissue penetrating the lobules proper. In lobules in which the central vein is intact it is surrounded by recent fibrous tissue which is connected with the periportal tissue by means of radiating strands. The amount of fibrous tissue varies greatly in the different lobules, but in all, the amount is relatively large. In many there is very little left but fibrous tissue, leucocytes and a few scattered liver cells. In others, perhaps one-third to one-half of the lobules are composed of hepatic cells grouped closely together (Fig. 3).

In the lobules, poor in liver cells, the cells are mostly pale and swollen. The cytoplasm is vacuolate, partly granular and partly hyaline. The nuclei are swollen and somewhat irregular in shape and staining. In some cells no nuclei can be found. In other words these cells are undergoing a peculiar form of degeneration ending in necrosis. In the larger cell-groups the liver cells are compact. Their cytoplasm is finely granular, staining well with eosin. Their nuclei are round, regular and take the nuclear stains well. A moderate number of these cells are binucleate and a few may be found with three nuclei. No definite mitotic figures are seen. The nucleolus is prominent and usually eccentrically placed. The chromatin appears to be somewhat more concentrated about the nuclear membrane and

about the nucleolus with a fine network connecting the two. The cells have the appearance of vigorous, regenerating cells.

In the periportal connective tissue are many proliferating "bile ducts." These appear as round or oval rings, or as long hollow tubes according to the way in which they happen to be cut. The epithelial cells lining them are packed closely together so that the nuclei appear to touch or actually to overlap one another. The cells are cuboidal in shape; their cytoplasm is basophilic, small in amount, and the cell boundaries are often indistinct. The nuclei are oval and stain deeply. No definite nucleolus can be made out. The chromatin is distributed as a fine network through the nucleus with somewhat thickened intersections.

The liver cells with their large amount of deeply staining cytoplasm, round nuclei, each with its nucleolus, are readily differentiated from the epithelial cells lining the "bile ducts," even though the two types of cells may be mixed quite indiscriminately. In many places small groups of liver cells which stain perfectly and which appear to be recently formed are scattered among the new "bile ducts" and are often arranged in the same form as the "ducts." Occasionally one sees a "bile duct" in which the cells seem to be undergoing transformation. At one end one sees typical bile duct epithelium, at the other end typical liver cells with cells in between which are not distinctly of either type.

Study of the circulatory apparatus shows large dilated veins in many of the periportal spaces with the compact groups of regenerating liver cells almost invariably located near these vessels. In many of the lobules the central vein cannot be found. In a number of places the bile capillaries are distended and filled with bile.

The whole picture, then, is one of a rather intense subacute inflammatory condition as shown by the edema, the large number of polymorphonuclear leucocytes and the widespread recent connective tissue proliferation. The simultaneous presence of marked destruction of the liver cells without evidence of pressure or other mechanical injury, suggests the direct action on them of some toxic substance. Both processes seem to run their course conjointly but more or less independently of each other, although the degenerative process in the liver tissue naturally facilitates the penetration of it by the connective tissue. There is also much evidence of liver cell regeneration.

The *spleen* shows a marked passive congestion with some thickening of the smaller arteries.

Bacteriologic examination. Smears from the liver contain a moderate number of neutrophilic leucocytes and a moderate number of short bacilli. Examination of sections of the liver stained with Giemsa fails to show any bacteria. Smears from the gallbladder contain no cells, many short bacilli. Culture of the bile shows many colonies of short gram-negative bacilli which are not motile, ferment glucose, coagulate milk, and show a heavy, brown growth on potato (*Bacillus coli*). Smears from the spleen show no bacteria.

Case 3

XXI. 108. E. S., an American housewife, 28 years of age. She entered the San Francisco Hospital with the complaint of pain in the stomach every morning, jaundice and general weakness.

The patient had had the usual diseases of childhood. She was divorced and had had no living children, but six abortions all of which had been induced. She had used beer to excess and had also used considerable whiskey for several years before her death. She had always led a strenuous life of dissipation. She had complained of stomach trouble, intermittently, for the past ten years, consisting of aching pains in the epigastrium in the mornings and after eating. She denied venereal infection.

The present illness had begun with jaundice which had come on suddenly four years before she entered the hospital. It had been associated with severe pains in the epigastrium. She had vomited several times and had passed blood in the stools. The jaundice cleared in four days but the pain persisted. Later hemorrhoids had appeared which at times disappeared for short intervals. The jaundice also had returned and had been complicated by diarrhea. She had lost 15 pounds in weight in six months. She also stated that she had coughed and vomited blood at times.

The clinical examination showed the abdomen distended and tight. The liver was enlarged and anastomoses between the epigastric and mammary veins were evident, but no *caput medusae* could be made out. The urine gave a strong reaction for bile. Blood examination revealed a marked secondary anemia and a slight leucocytosis. The hemoglobin content was 60 per cent. The red cells numbered 2,600,000 per c.mm., the white cells 14,000 with 80 per cent polymorphonuclear neutrophiles.

Clinical diagnoses. Portal cirrhosis; parenchymatous nephritis; aortic stenosis.

Anatomic diagnoses. Alcoholism; cirrhosis of the liver; fatty liver; gastritis, chronic; enteritis, acute; broncho-pneumonia, terminal; salpingitis, chronic; appendicitis, chronic; peritonitis, healed; perisplenitis, chronic; pancreatitis, chronic; jaundice; anemia, secondary.

Abstract of the autopsy record. The body is that of a strongly built woman. The abdomen is slightly distended. The sclerae are icteric; jaundice is marked over the entire body. The peritoneum is smooth. The liver is large, the lower border extending 13 cm. below the xiphoid process in the midline. It extends also fairly well to the left while on the right side it is midway between the costal margin and the crest of the ilium. About 250 c.c. of clear fluid containing some flakes of fibrin are found in the peritoneal cavity. The diaphragm level is at the 4th interspace on both sides. The heart is enlarged slightly, otherwise nor-

mal. The lungs are edematous with beginning broncho-pneumonia at their bases. The spleen is somewhat enlarged (14 x 9 x 6 cm.). The capsule is strongly adherent to the abdominal wall and to the omentum. The kidneys are somewhat swollen and the cut surface is of a grayish-yellow color. The mucous membrane of the stomach is markedly congested and shows a few hemorrhages.

The *liver* is distinctly enlarged measuring 33 x 27 x 10.5 cm. There are fibrinous adhesions over the entire surface of the capsule. The liver is yellow, has a nodular appearance and feels very firm under the knife. The cut surface also shows a nodular appearance with considerable scarring between the nodules.

Histologic examination.

Liver. There are two very striking things in the microscopical appearance of this liver; first, the extreme amount of fatty infiltration; second, the diffuse development of the connective tissue. The latter is greatly increased about the periportal spaces and shows numerous arborizations of finer branches which dissect the lobules or pseudo-lobules producing thereby a very diffuse intralobular fibrosis. As would be expected, the larger cell-groups differ markedly from the true lobules. They vary considerably in size but usually do not attain the size of a normal liver-cell unit. A few lobules show a moderate fibrosis about the centers giving the appearance of a wheel with hub and radiating spokes composed of fibrous tissue (Fig. 5). The trabecular arrangement of the liver cells is almost gone, the cells apparently accommodating themselves to the irregular spaces in the meshwork of connective tissue. The latter is quite cellular and fairly recent. The fibrous tissue is moderately, diffusely infiltrated with round cells; among them are a good many polymorphonuclear leucocytes. In many places small groups of liver cells in the meshes of the new connective tissue show hyaline degeneration and have vacuolate, coarsely granular cytoplasm. They have swollen, pale-staining or very often shrunken nuclei. The process evidently ends in necrosis.

The fatty infiltration is very general. On the other hand in the small groups, or in the periphery of some of the larger ones, are cells with little or no fat. These appear to be normal, active cells. They are relatively few in number and exhibit little tendency to proliferation.

Proliferation of the "bile ducts" is not an outstanding feature of this liver but they are present in a moderate number. Here, as in the preceding cases, apparent transformations between "ducts" and rows of liver cells can be found but they are by no means so plentiful

or so evident as in Cases 2 and 4. There are a few liver cells scattered among the proliferating "ducts" which look like recently formed cells.

The picture presented in this case varies from that in the two preceding cases in the presence of an extreme fatty infiltration and in the absence of any clear evidence of regeneration in the liver cells. In common with the others are the extensive fibrosis, the marked cellular infiltration and the apparently more or less independent, but nevertheless quite general, destruction of liver cells.

Examination of sections of the liver stained with Giemsa fails to show any bacteria.

The *spleen* shows marked passive congestion and some fibrous thickening of the capsule. In the *pancreas* a moderate diffuse, interstitial fibrosis is present with some areas of fat necrosis. The mucous membrane of the *stomach* is moderately infiltrated with round cells and somewhat shrunken in places.

Case 4

XIX. 48. B. C., an Italian dishwasher, a married man of 48 years, entered Lane Hospital complaining of severe pains in his arms and legs, which had lasted for one month.

His general health in the past had been good. He denied venereal infection. He had used alcohol to great excess, stating that he had been drunk most of the time when not working.

The present illness commenced suddenly, one month before entry to the hospital, with pains in the legs and swelling which a week later extended to the arms and the back. His mentality was not clear.

Clinical examination revealed a strongly built, slightly obese man sleeping most of the time. The muscles showed occasional twitchings. Signs of pneumonia were noticed in both lungs. The liver edge was not palpable but there was dullness for 15 cm. below the margin of the ribs in the right mid-clavicular line. Partially healed ulcers and pigmented scars were found on both legs. Both legs were edematous.

The blood picture was that of a moderate secondary anemia with marked anisocytosis and slight poikilocytosis. An occasional nucleated red cell was found in the smears. The hemoglobin content was 84 per cent (Sahli). The red cells numbered 3,100,000 per c.mm., the white cells 10,000. The differential count was normal. The Wassermann reaction was negative in the blood and spinal fluid. A blood culture showed no growth.

While in the hospital the patient showed increasing mental symptoms, arising at night and wandering about the ward resenting any restraint from attendants.

Clinical diagnoses. Delirium, alcoholic; broncho-pneumonia; pyorrhea alveolaris; arteriosclerosis, general with arteriosclerotic kidneys; paraphimosis.

Anatomic diagnoses. Tuberculosis of the lungs, chronic; tuberculosis of the intestines; broncho-pneumonia; pleurisy, chronic; pleurisy, acute; alcoholism, chronic; cirrhosis; balanoposthitis; cystitis, chronic; pyelitis, chronic; pyelonephritis, acute; prostatitis, chronic; conjunctivitis; emphysema.

Abstract of autopsy record. The body is that of a strongly built, muscular, well-nourished man. The conjunctivae are slightly yellow. The feet are somewhat edematous. The abdomen is moderately distended. The peritoneum shows slight passive congestion, but no free fluid is present in the peritoneal cavity. The chest is distinctly barrel-shaped. There are two old cavities in the upper lobe of the right lung. There are also some scattered areas of gray consolidation and some recent tubercles in both lungs. The spleen is slightly enlarged, measuring 17 x 8 x 2.5 cm. Its cut surface is dark red and soft. The gallbladder and the bile ducts are normal.

The *liver* is rather large measuring 30 x 20 x 8 cm. Its tissue is very firm and cuts with considerable difficulty. The cut surface shows numerous small bright yellow spots with gray lines between them. The tissue is especially indurated between the right and left lobes.

Many tuberculous ulcerations are present in the small and large intestines.

Histologic examination.

Liver. The fibrosis in this liver is extensive and in general of the multilobular type. The parenchyma is cut up into irregular patches by the broad bands of fairly old connective tissue (Fig. 7). Throughout the latter one finds a marked cellular infiltration consisting of lymphocytes, a few polymorphonuclear leucocytes and some eosinophiles (Fig. 8). The islands of liver tissue surrounded by this widespread development of connective tissue vary in size. The smaller ones are one-fourth to one-fifth the size of a normal lobule. The larger ones include perhaps two or three lobules or parts of several lobules. The latter are not numerous, however, and usually finer strands of connective tissue dissect these into blocks somewhat smaller than the ordinary hepatic unit.

The liver cells show a moderate fatty infiltration which is quite generally distributed through the sections (Fig. 7). The structure of the lobules is greatly disturbed. The cells have the appearance of being crowded or packed, without any definite arrangement, into the spaces formed by the bands of fibrous tissue. The radial trabecular structure is lacking. The central vein is absent in many lobules and eccentrically placed in others.

Many cells in the smaller and larger groups, and small isolated cell-groups caught in the dense fibrous bands, show hyaline degeneration of the cytoplasm. In others the cytoplasm is swollen, coarsely granular and vacuolated. The nuclei often stain very deeply. Others are swollen, pale, irregular in shape; again others have disappeared completely indicating that many of the cells are undergoing necrosis (Fig. 8).

By far the greater number of cells, however, appear well filled out. Their cytoplasm is finely granular and stains evenly. Their nuclei are round and contain chromatin of normal distribution and staining quality. These are presumably active, functioning cells. Many groups seem to show evidence of regenerative proliferation. Some of these latter cells have large round nuclei and others have two nuclei. While no definite mitoses can be found many of these large nuclei appear to be entering the prophase.

Many proliferating "bile ducts" can be found in and along the edges of the fibrous tissue. The relation of them to the liver cells is interesting. As in Case 2, many seem to be undergoing transformation and various stages may be found between the true "bile-duct" epithelium and typical liver cells.

There is also evident in this case, as in the second one, a certain amount of edema which is discernible in the lobules in places where the cells are not so closely packed.

The general picture in this case seems to be that of an inflammatory condition perhaps of longer duration but less severe than that in the second case. The widespread production of fibrous tissue, the presence in it of cellular infiltration including polymorphonuclear leucocytes, and the presence of edematous fluid all emphasize the distinctly inflammatory character of the lesions in association with a probably toxic degeneration and necrosis of large numbers of liver cells followed by regeneration. The evidence, furthermore, indicates that the process is still progressive.

Examination of sections of the liver stained with Giemsa fails to reveal any bacteria.

Comment

All four patients had taken alcohol to excess for a considerable period of time and two had died with the symptoms of an alcoholic psychosis. Although a discussion of the etiology of cirrhosis is beyond the scope of this paper, we may state in passing that we regard the toxic effect of alcohol as one of the causal factors in this type of cirrhosis. Experimental work, however, points strongly to the existence of other factors which may play an equally essential rôle. The nature of these accessory factors is at present unknown.

Rolleston [5] states that cirrhosis is usually fatal at about 50 years of age. Our cases, as would be expected, since in them the lesions

are relatively early, ended in death at an earlier age than this, the average being 38 years. In the different patients death had occurred at the ages of 39, 38, 28 and 48 years respectively. In none of our cases is death directly chargeable to cirrhosis of the liver, a finding which conforms to Hawkins'[2] observation that early cirrhosis is seen only when death occurs from some other cause. In Case 1 death was due to the rupture of a syphilitic aneurism; in Cases 2 and 3, death was the result, primarily, of the general alcoholic intoxication; while in Case 4 it was caused by the combined effects of tuberculosis (pulmonary and intestinal), broncho-pneumonia and alcoholism.

While most modern writers on cirrhosis are fairly well agreed that the primary lesion in cirrhosis consists in a degeneration of the liver cells which is followed by connective tissue proliferation, our cases seem to show that from the very beginning these two processes occur conjointly and simultaneously. The very early lesions in the liver tissue in Case 1 illustrate this very well. Here we find small foci of liver cell degeneration together with a definitely inflammatory proliferation of the connective tissue (Fig. 2). The inflammatory character of the changes is made evident by the presence of an inflammatory exudate containing polymorphonuclear leucocytes and lymphocytes. In these areas the connective tissue cells are proliferating quite diffusely without close topical relation to the degenerating cells although the advance of the proliferating connective tissue into the liver tissue is evidently favored by the degeneration and necrosis of the liver cells. In our opinion, such lesions as these can be best explained by assuming that the toxin or toxins which injure the hepatic cells in such a way as to cause degeneration ending in necrosis, probably at the same time have an irritating effect upon the more hardy connective tissue, stimulating the connective tissue cells to growth and multiplication and the building up of new fibrous tissue. The reaction would be equivalent to that which occurs so commonly in other forms of proliferative inflammation of infectious or toxic origin.

The distinctly inflammatory nature of the lesions is very evident in all four of our cases. In all of them not only lymphocytes but also polymorphonuclear neutrophilic leucocytes are found in the proliferating connective tissue and about the degenerating liver cells. These are most plentiful in Cases 1 and 2, which are more acute than

the others (see Figs. 2 and 4). In Case 2 they are the predominating type of leucocyte. In addition, signs of an inflammatory edema among the degenerating liver cells and in the new connective tissue are quite evident in Cases 2 and 4.

Leucocytic infiltration, edema and proliferating fibroblasts form the inflammatory part of the process which, however, would not be complete or lead to so much destruction if it were not associated with the degenerative process in the liver cells. The importance of the latter is also clearly shown in all our cases whether they are more acute or more chronic. In all of them microscopic examination reveals much injury to the liver cells resulting in swelling, hyaline and granular degeneration of the cytoplasm, vacuolization and nuclear changes ending in the total disappearance of the nucleus.

The new connective tissue in our cases varies in age from the early fibroblastic stage, in Cases 1, 2 and 4 (Figs. 2, 4 and 8), to that of a fairly cellular fibrous tissue in Cases 3 and 4 (Figs. 5 and 7), but we believe that all four cases still belong to the group of subacute cirrhosis.

Even in these relatively early cases there is abundant evidence of regenerative proliferation of the liver cells. In Case 1 active multiplication of them is clearly shown by the crowding of large numbers of well-preserved cells into the lobules in an irregular fashion (Fig. 1). In Cases 2 and 4 (Figs. 3 and 7) the regenerating cells are found in groups which are more or less circumscribed and which are the forerunners of the hyperplastic nodules so familiar in the hob-nail liver (MacCallum).[6] In our cases these agglomerations are not so large as the nodules commonly found in the late stages of the disease, and since the fibrous tissue around them has not yet contracted, they are less prominent. This explains the relatively smooth surface of the liver in our cases. The evidence of regeneration is less clear in Case 3 than in any of the others.

Many of the earlier writers on cirrhosis have called attention to the increase in the size of the liver in the early stages of the disease. In all four of our cases the liver is above normal size. In Case 1, a woman, the liver is moderately enlarged, measuring 26 x 26 x 7 cm. In Case 3, also a woman, the liver is distinctly increased in size, measuring 33 x 27 x 10.5 cm., while in Cases 2 and 4, who are men, there is a moderate enlargement of the organ, the measurements being 27 x 22 x 10 cm. and 30 x 20 x 8 cm., respectively. Although

there is marked destruction of the liver tissue in all four cases, there are several factors which tend to offset this and which may account for the increased bulk of the liver. These factors are: (1) the increase in young connective tissue; (2) the leucocytic infiltration; (3) the presence of edema; (4) regeneration of liver cells.

Kretz[7] has demonstrated clearly, by means of injected specimens, the profound changes in the blood vessels of the liver which occur in portal cirrhosis. He finds the portal vein dilated, its walls thickened, and its ramifications more numerous than in the normal liver. We find that these changes are fairly well marked even in these early cases. The portal vein is often dilated and its branches are more numerous than in the normal liver. Case 2, which is one of the earlier cases, shows this quite well. On the other hand, according to Kretz, the hepatic veins are reduced in number. This also coincides with our own observations. In many lobules they cannot be found at all while in others they are eccentrically placed.

The development of bile duct-like structures is especially well marked in two of our cases. The relationship of these new "ducts" to the liver cells is a very interesting one, but the controversy over the part which they play in the regeneration of liver tissue is still to be decided.

When, as pointed out in the records, the proliferating "bile-ducts" are occasionally seen to end in club-shaped groups of liver cells with a transitional type of cell connecting them, it is natural to assume that the "bile ducts" are undergoing transformation into rows of liver cells. This assumption is strengthened by the presence of regenerated liver cells in the vicinity; and by the fact that embryologically the liver cells develop from the epithelium of the primitive bile ducts. The proliferating "ducts" are usually found in the new connective tissue often near or scattered among hepatic cells which have the appearance of being recently formed. Furthermore, the "ducts" are not found in the regions where the liver cells are undergoing destruction. These observations suggest that the "bile ducts" function in the process of regeneration. The appearances pointing to the transformation of "bile ducts" into liver cells is very marked in Cases 2 and 4. In Case 2, especially, many "bile ducts" can be found which terminate in rows of liver cells which have every appearance of being recently formed. Our findings coincide with the observations of many of the prominent investigators of regeneration

of the liver such as Waldeyer (1868), Hanot (1895), Stroebe (1897), Rolleston (1905), Meder (1905), and MacCallum (1904). It may be pointed out however that such interpretation of the development of one type of cell into another from the presence of "transitional" stages is always open to criticism.

On the other hand, some of the "bile ducts" are narrow, flattened strands of cells which appear to have been mechanically compressed. These evidently show that some of the "bile ducts" arise from atrophied rows of liver cells rather than the reverse.

Conclusions

1. Histologic study of relatively early, progressive cases of alcoholic cirrhosis reveals the inflammatory nature of the changes in the connective tissue by the presence of the usual signs of a subacute proliferative inflammation, namely, inflammatory edema, infiltration with leucocytes, among them many polymorphonuclear neutrophiles, and proliferation of the connective tissue cells.

2. Evidence of injury to the liver cells is always present but apparently does not precede the proliferation of the connective tissue. The two processes occur simultaneously, probably as a result of the action of a common cause.

3. The characteristic hyaline degeneration of the cytoplasm of the hepatic cells, pointed out by Mallory as peculiar to alcoholic cirrhosis, was found in all our cases.

4. The history of all four patients revealed chronic alcoholism and alcoholism was the principal cause of death in two of them.

5. Regeneration of liver cells may be a prominent feature even in early cirrhosis.

6. In all our cases the liver was somewhat larger than normal, in one of them it was considerably enlarged.

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DESCRIPTION OF PLATES LXXVIII–LXXXI

FIG. 1 (Case 1). Low power view of the liver showing patchy distribution of the connective tissue, cellular infiltration, fatty infiltration and pale-staining liver cells (glycogen). The cells are crowded and the normal arrangement of the lobule is disturbed due to rapid regeneration of liver cells.

FIG. 2 (Case 1). High power view of early focal lesion, showing destruction of liver cells with hyaline and granular degeneration of the cytoplasm, pyknosis and disappearance of nuclei, marked infiltration with polymorphonuclear leucocytes and lymphocytes, active proliferation of connective tissue cells.

FIG. 3 (Case 2). Low power view showing widespread destruction of liver tissue, extensive production of new connective tissue and marked cellular infiltration, many new "bile ducts" in the connective tissue. At the lower edge is a group of regenerating liver cells.

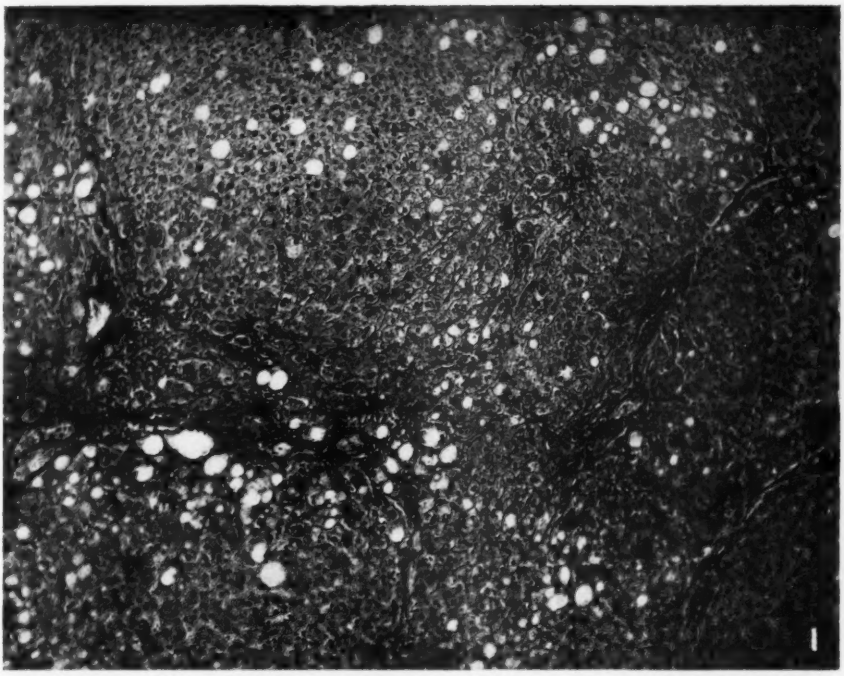
FIG. 4 (Case 2). High power view showing destruction of liver cells. The cells are swollen, several show hyaline and granular degeneration of the cytoplasm with marked vacuolization in some. Several cells are almost completely destroyed. The nuclei of the degenerated cells are shrunken, pyknotic, some have completely disappeared. The new connective tissue is heavily infiltrated with polymorphonuclear leucocytes.

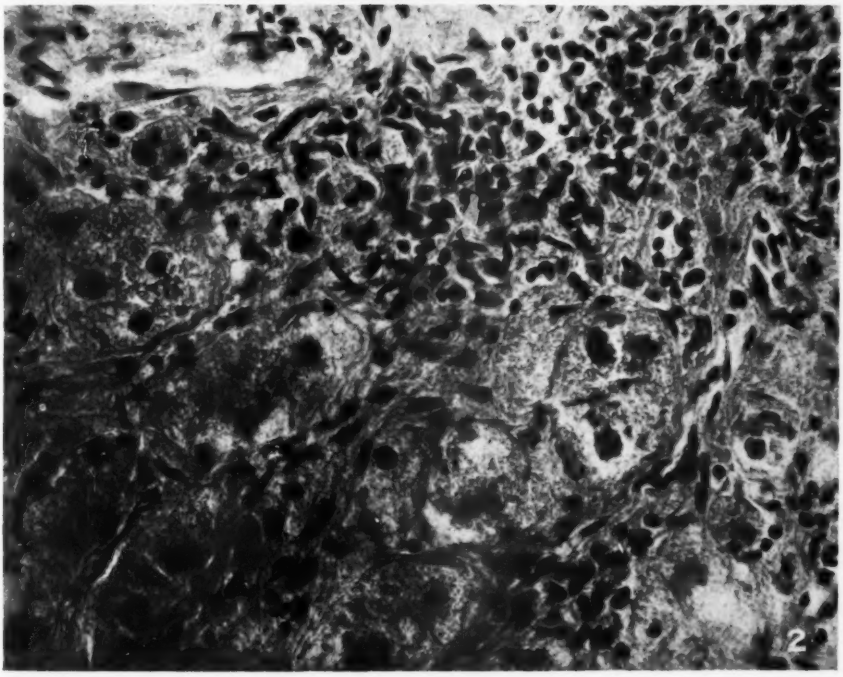
FIG. 5 (Case 3). Low power view showing the peculiar distribution of the new connective tissue and the extensive fatty infiltration of the liver cells, also the marked cellular infiltration of the fibrous tissue.

FIG. 6 (Case 3). High power view showing marked destruction of liver cells with granular and hyaline degeneration and vacuolization of the cytoplasm. Several cells show large globules of fat in the cytoplasm. Proliferation of the connective tissue can be seen among the degenerating cells; also infiltration with polymorphonuclear leucocytes and lymphocytes.

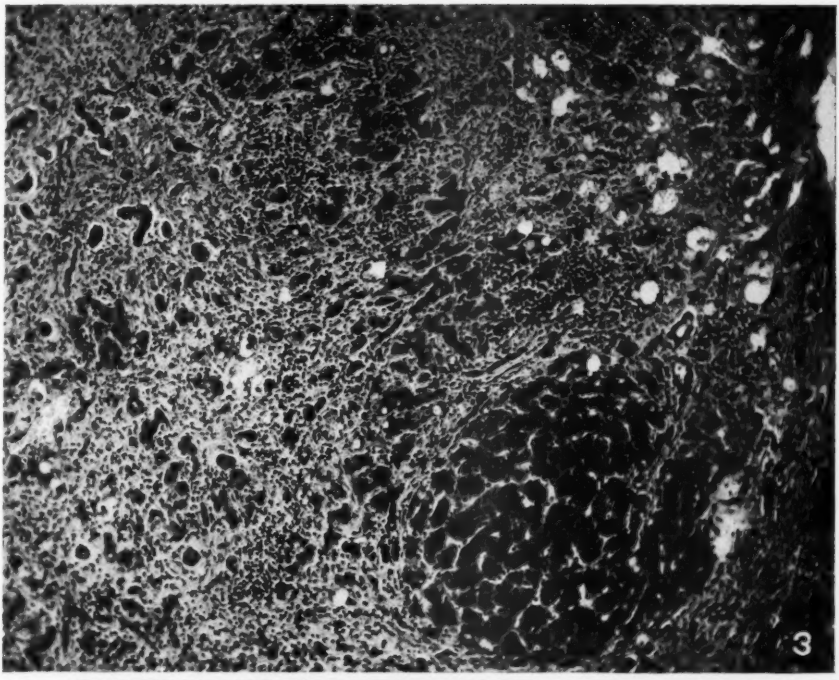
FIG. 7 (Case 4). Low power view showing distribution of the abundant fibrous tissue with very heavy cellular infiltration. There is some fatty infiltration and much evidence of regeneration of liver cells.

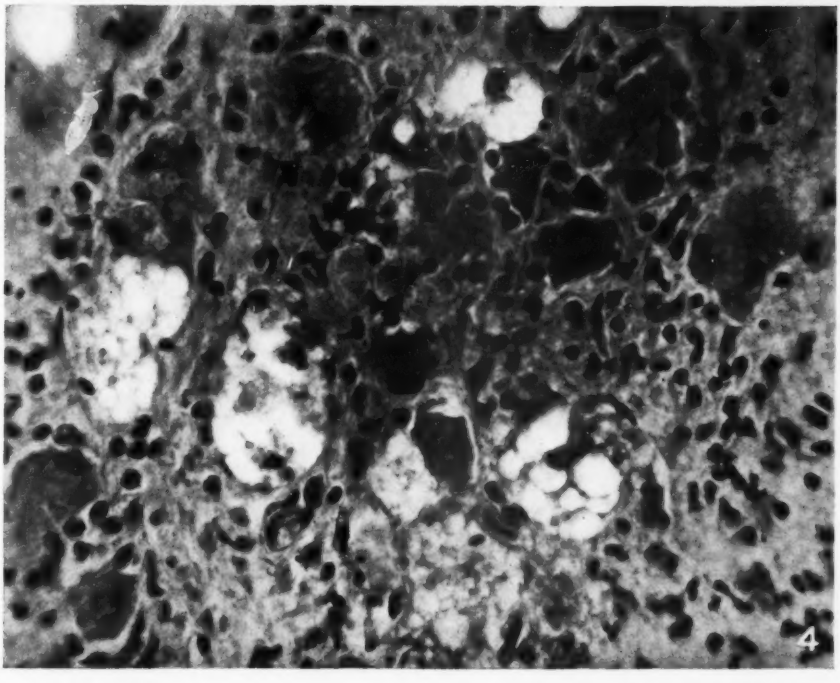
FIG. 8 (Case 4). High power view showing destruction of the liver cells with extensive hyaline and granular degeneration of the cytoplasm. The new connective tissue is heavily infiltrated with polymorphonuclear leucocytes and lymphocytes.



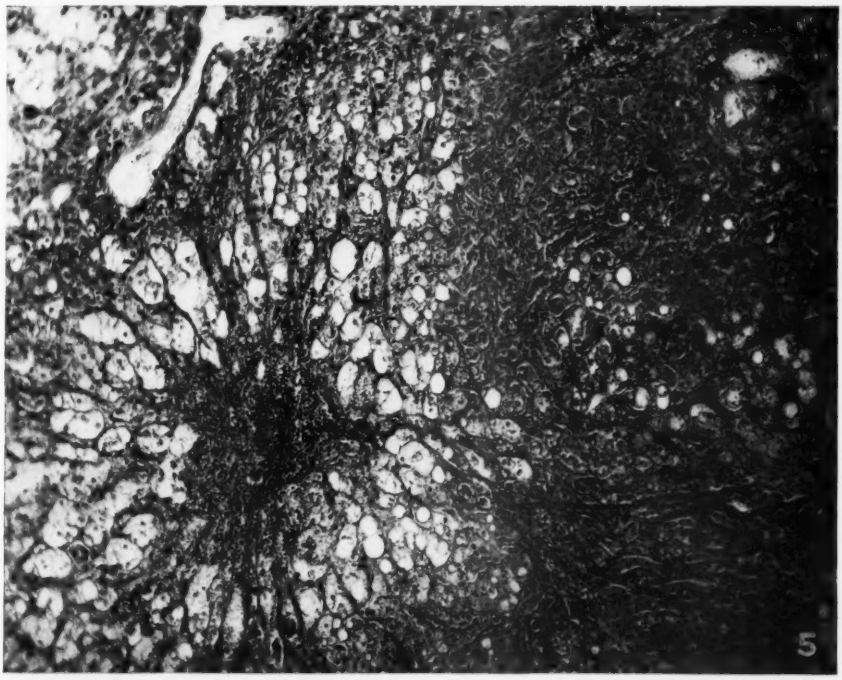


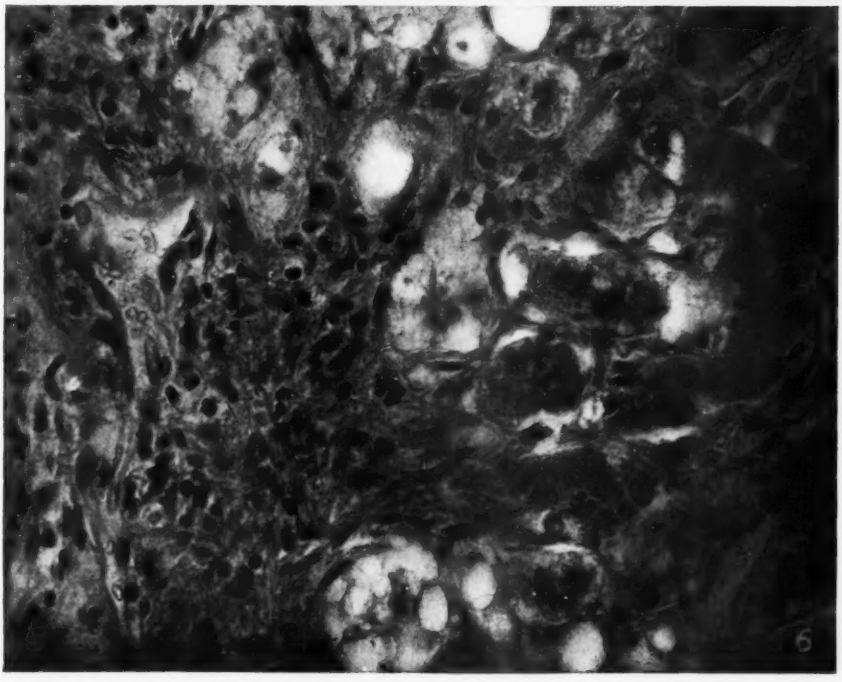
Hall and Ophüls Progressive Alcoholic Cirrhosis



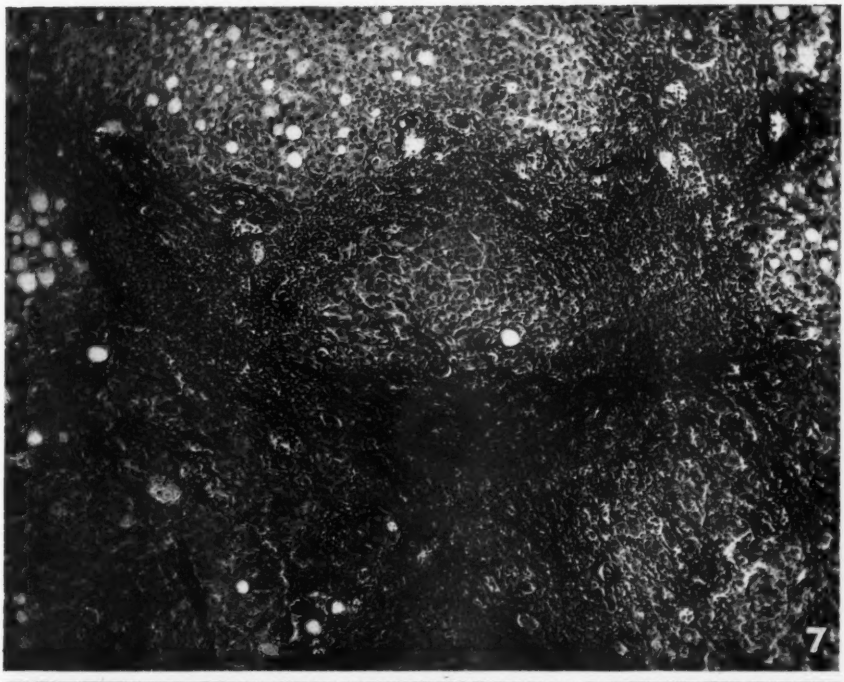


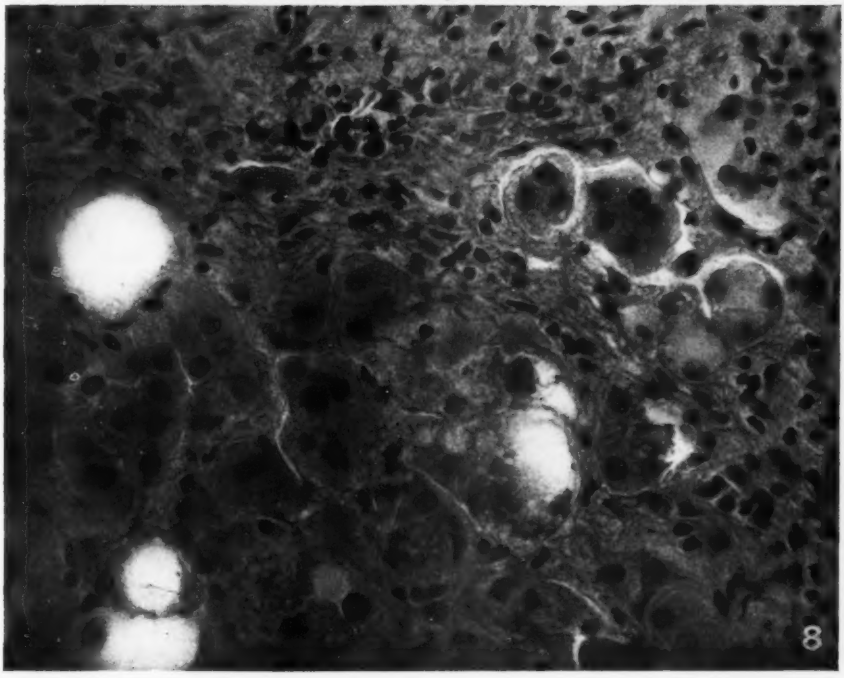
Hall and Ophüls Progressive Alcoholic Cirrhosis





Hall and Ophüls Progressive Alcoholic Cirrhosis





Hall and Ophüls Progressive Alcoholic Cirrhosis

OBSERVATIONS ON FOCAL TUBERCULIN HYPERSENSITIVENESS IN AN INFECTED ORGAN PREVIOUS TO A GENERAL SENSITIZATION *

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In a previous study [1] concerned with allergic reactions in testes of guinea-pigs sensitized by treatment with killed tubercle bacilli, the observation was made that definite attempts at healing occurred in testes of untreated control animals before these animals had become generally hypersensitive. By "healing" is meant especially fibroblastic proliferation, though of course healing in tuberculosis includes much more than this. The untreated control animal reacted to testicular infection with the B–I strain of tubercle bacillus with fibrosis before the pig had become generally sensitized by his infection or at least before an intradermal tuberculin reaction had been positive.

This fact raised a question as to whether or not an animal with testicular infection would react to tuberculin introduced into the infected organ before the animal gave a positive intradermal reaction to like dilution; in other words, is a local sensitivity to tuberculin demonstrable in an infected organ before fibroblastic proliferation, as evidence of walling-off or "healing," becomes evident? To test this question the following experiment was devised. Seven normal guinea-pigs, each weighing over 600 gm., were given 0.1 c.c. of a saline suspension containing 2.5 mgm. per c.c. of living bovine tubercle bacilli. The B–I strain was used and the right testis infected. On the second, third, fourth, fifth, sixth, seventh and eleventh day after infection, 0.1 c.c. of 5 per cent old tuberculin was injected both into the infected and into the normal testis, and a like quantity was given intradermally. Animals were killed about eighteen hours after testing. In a second series of five pigs both testes were infected with similar doses of the same bovine strain; two, four, six, seven and eight days later one testis was tested with 5 per cent tuberculin, the intradermal test was done, and the second testis received 0.1 c.c.

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of physiological saline. In a single uninfected animal, one testis was injected with 5 per cent tuberculin, the other with physiological saline. Again, animals were killed eighteen hours after testing. Testes were fixed in Zenker's fluid, cut at 6 microns and stained in eosin-methylene blue. Some were fixed in alcohol-formalin, embedded in celloidin and stained with hematoxylin and eosin.

It may be stated at the outset that definite focal reactions to tuberculin occurred in the infected organ before a positive skin test was obtained. This brings up the question as to what constitutes a positive intradermal reaction. Tuberculin is not innocuous to the normal animal. The author has read upwards of 1000 intradermal tests on guinea-pigs and has frequently observed in normal animals transient redness or transient induration persisting twenty-four to forty-eight hours after intradermal tuberculin and has occasionally carefully autopsied an animal giving a questionable positive skin test, always with negative results as to the presence of infection. Simple blanching of the skin likewise is common, possibly being the rule in an unsensitized animal. The combination, however, of central blanching, with surrounding erythema and induration, even if slight, is always significant, and in the writer's recollection has almost always meant sensitization or infection verifiable either by autopsy or by other methods. One exception to this rule was the case of a female guinea-pig confined with a male sensitized by killed tubercle bacilli injected into the testis. This pig gave several very suggestive reactions, with blanching, induration and erythema but sensitization was never verified; there have been other rare instances. In the present study a "positive" intradermal reaction refers to one with at least central blanching, peripheral erythema and induration.

Some reaction is produced by injection of 5 per cent old tuberculin in the testis of a normal guinea-pig. Eighteen hours after such injection histologic examination shows essentially normal tubules, with slight evidence of desquamation or cessation of spermatogenetic activity. Between the tubules, however, there is a moderate fibrinous exudate (Fig. 6) and numerous polymorphonuclear leucocytes and eosinophiles are present. Lymphocytes are rare but a moderate number of phagocytic endothelial leucocytes occur. These latter seem highly phagocytic for polymorphonuclears. Changes induced by tuberculin in the normal testis are diffuse and affect the entire organ.

Physiological saline in the normal testis produces no definite alteration within eighteen hours, although an occasional extravascular lymphocyte may be found. Traces of fibrin present are probably due to trauma.

The first pig of Series I, infected in the right testis, received 0.1 c.c. of 5 per cent old tuberculin, forty-eight hours later, in each testis, and a like quantity intradermally. Eighteen hours afterward the intradermal test was negative. Grossly the right testis was more injected than the control and the tunicae were non-irritable; that is to say, the fine tremors seen in the muscle after death and exposure to air currents were lacking. Microscopically, however, aside from a small extravasation of blood at the point of entry of the needle, there was nothing beyond what might be expected from tuberculin injected in a normal testis, save for a certain increased amount of desquamation of epithelium and infiltration by endothelials, easily explainable on the basis of infection alone. At least it may be stated that there was no focal change such as we shall see occurs after tuberculin in animals subsequently described. The opposite uninfected testis shows a polymorphonuclear infiltration and traces of desquamation not exceeding that seen in testes of normal tuberculin-injected guinea-pigs. There was no focal lesion. In Pig II, tested in the same manner as the preceding, but twenty-four hours later, in addition to polymorphonuclear infiltration, and the usual evidences of tuberculous infection, there are a small number of tubules which show much more marked desquamation, actual necrosis and extensive invasion of the epithelium and of the tubular lumen by polymorphonuclears. In addition there is considerable subcapsular fibrin, some hemorrhage possibly due to the trauma of injection, and much polymorphonuclear reaction beneath the capsule. In other words there are focal changes; their significance is only determinable in the light of later stages. The skin test and the opposite tuberculin-injected testis of Pig II were not remarkable.

Pig III received tuberculin into each testis and intradermally four days after testicular infection. Eighteen hours later the skin showed a slight atypical blanching with neither surrounding erythema nor induration, a test interpreted as a frank negative. The control testis exhibits possibly a trace more reaction than one might expect in an unsensitized animal but too little to be certain. Changes likewise are not focal. The infected testis, however, in addition to

the general changes due to infection shows a well-marked focus of increased desquamation and necrosis of tubules with marked surrounding polymorphonuclear infiltration and invasion of pink-staining necrotic tubules by polymorphonuclear leucocytes. These changes are certainly to be interpreted as reaction. Unfortunately the sections are from celloidin-embedded material and good photographs could not be obtained. Pig IV received identical treatment five days after infection, and was killed on the sixth day. The skin showed simply blanching without erythema or induration. The control testis microscopically is negative save for the non-specific tuberculin reaction (Fig. 5) and evidences of reaction to the trauma of needle puncture. Figure 1 gives some idea of the histology in the infected tuberculin tested testis. The lower half of the figure, approximately, shows the appearance of the infected testis. The tubules show no evidence of spermatogenesis. The epithelium is desquamated and elsewhere in the organ the lumina of tubules contain large epithelial giant cells formed either by fusion or multiple division of epithelial cells. Other tubules show large vacuolated cells. Surrounding these tubules the reaction is mainly of the endothelial and lymphocytic type (Fig. 7). Polymorphonuclear leucocytes and eosinophiles are rare. There is a heavy deposit of fibrin in the subcapsular region and between testis and epididymis, and large numbers of extravascular red cells are present. The upper portion of the figure possesses an entirely different appearance. The tubules exhibit a degree of necrosis far beyond a possibility of adequate demonstration by photomicrograph. Epithelial cells are almost wholly desquamated (Fig. 4); they take a deep pink eosin stain characteristic of necrotic tissue. Nuclei are largely absent and surrounding almost every tubule are dense collections of polymorphonuclear leucocytes. Curiously enough in these early tuberculin reactions edema is lacking or is present only in traces. Grossly the infected testis of Pig IV was no larger than the control and neither was larger than the normal organ. The infected testis differed grossly from the control only in degree of redness, the size of dilated venules and in the appearance of the tunicae; the latter in the infected testis were loosely adherent to the organ. In all early stages necrosis is the marked distinguishing feature, and had it not been for histologic examination this would not have been apparent and the testis would probably have been noted as non-reacting.

Pig V, tuberculin-injected as were the preceding, on the sixth day and killed eighteen hours later showed a negative intradermal test. In the infected testis, however, the necrotic tissue was very evident in gross; the testis presented an area of marked pallor with surrounding erythema resembling features characteristic of an early positive intradermal test. The control testis was not remarkable. Histologically (Fig. 2) the infected testis resembles that of Pig IV, a typical progressive tuberculosis with much endothelial cell infiltration, lymphocytes, plasma cells and rare polymorphonuclears. Beside this there is a sharply outlined necrotic zone, with pink-staining tubules filled with non-nucleated, desquamated epithelium and surrounded by dense masses of polymorphonuclears. For the first time the control testis shows evidence of reaction, similar to that found in the infected testis. There is a very minute focus where typical necrosis of tubules with surrounding polymorphonuclear leucocytes stamps the organ as a reacting structure. Edema is absent.

Pig VI, twenty-four hours later, had a negative intradermal test. The infected testis reacted similarly to the testes of earlier animals, and its histologic appearance (Fig. 3) is almost identical with them. The uninfected control testis shows a small focal reaction, no more than did the previous control.

The remaining animal was kept until the eleventh day after infection before being similarly treated with tuberculin. It was felt necessary to be certain that a positive intradermal test would eventually be obtained. Eighteen hours after the usual injection the intradermal test was definitely positive although not a strong positive. There was central blanching with a faint bluish tint, edema, surrounding erythema and induration, but no eschar. Both testes, however, were enlarged to twice normal size, were reddened, firm and edematous. The spermatic cord was injected and edematous and there was blood-tinged fluid in the peritoneal cavity. Liver, spleen and retroperitoneal nodes showed tubercles. Microscopically both testes exhibit the usual typical reactions of the sensitized testis, — necrosis, polymorphonuclear infiltration and here for the first time there is extensive edema.

The second series of pigs was used to eliminate the factor of trauma, since it was not certain that the necrosis observed after tuberculin injection in an infected organ was not traumatic. Therefore,

both testes were infected each with the same dosage of organisms as that used in Series I, doubling consequently the total infecting dose. Then at stated intervals, one testis and the skin received each 0.1 c.c. of 5 per cent old tuberculin and the second testis was injected with 0.1 c.c. physiological saline. Forty-eight hours after infection the first pig was treated in the fashion just described. The skin test was negative, the saline-injected testis is negative histologically save for the early evidences of infection and the tuberculin-injected testis shows no change which may be interpreted as due to sensitization. The second animal, tuberculin-treated at ninety-six hours and killed eighteen hours later, gave a negative skin test and its testes show no focal changes microscopically save those of infection. At 144 hours the pig showed a skin test interpreted as a weak positive; the saline-injected testis shows no reaction histologically but the tuberculin treated testis exhibits a weak reaction. This pig has been the one exception in the two series and it may be that in some manner it received a larger infecting dose and reacted generally earlier than did others of the series. Or, more likely, since the focal testicular reaction is weak, it may be that the intradermal test was one of the rare false positives. The two remaining animals of Series II injected at 168 and 192 hours both gave negative intradermal reactions but reacted focally in the infected testis in the same manner as did animals of the first series.

Conclusions

1. The guinea-pig testis infected with bovine tubercle bacilli reacts focally to 5 per cent old tuberculin before the opposite uninfected testis shows a positive reaction and before a positive intradermal test is demonstrable.

2. This focal reaction is characterized by a sharply demarcated zone of increased necrosis of tubules surrounded by a dense exudate of polymorphonuclear leucocytes.

3. Physiological saline in an infected testis will not give a similar picture.

4. The significance of this earlier focal hypersensitiveness in an infected organ is wholly problematical. It probably means merely a more intense local flooding with antigen, but can have no bearing on the question of local origin of antibodies or of local immunity.

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DESCRIPTION OF PLATES LXXXII-LXXXV

Fig. 1. Infected testis, tuberculin-injected on the fifth day. Necrotic tubules occupying approximately the upper half of the figure. Dense exudate of polymorphonuclears surrounding the necrotic tubules. X 100.

Fig. 2. Infected testis, tuberculin-injected on the sixth day. Necrotic tubules in the upper half of the figure, polymorphonuclears forming a sharp boundary between reacting and non-reacting portions of the testis. X 100.

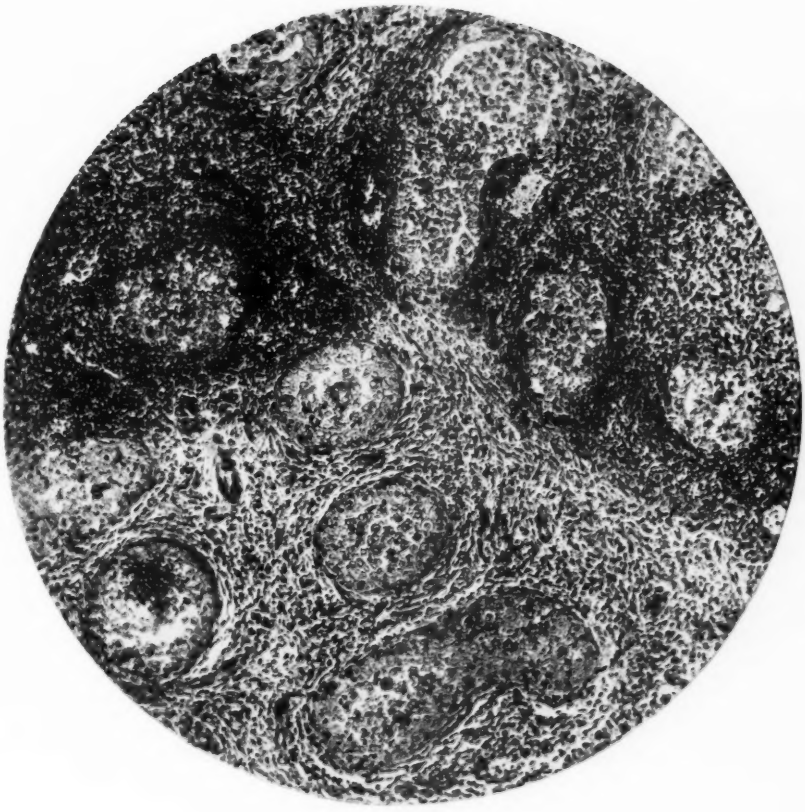
Fig. 3. Infected testis, tuberculin-injected on the seventh day. The usual sharp line of polymorphonuclears separating necrotic from non-necrotic tubules. X 100.

Fig. 4. Necrotic tubules and polymorphonuclear reaction in infected testis, tuberculin-injected on the fifth day. X 250.

Fig. 5. The opposite uninfected testis of the same animal. This testis received the same dosage of tuberculin. X 200.

Fig. 6. Showing the effect of tuberculin in the testis of an uninfected animal. X 200.

Fig. 7. Infection alone: the zone outside of the reacting tissue, infected testis, tuberculin-injected on the fifth day. X 250.



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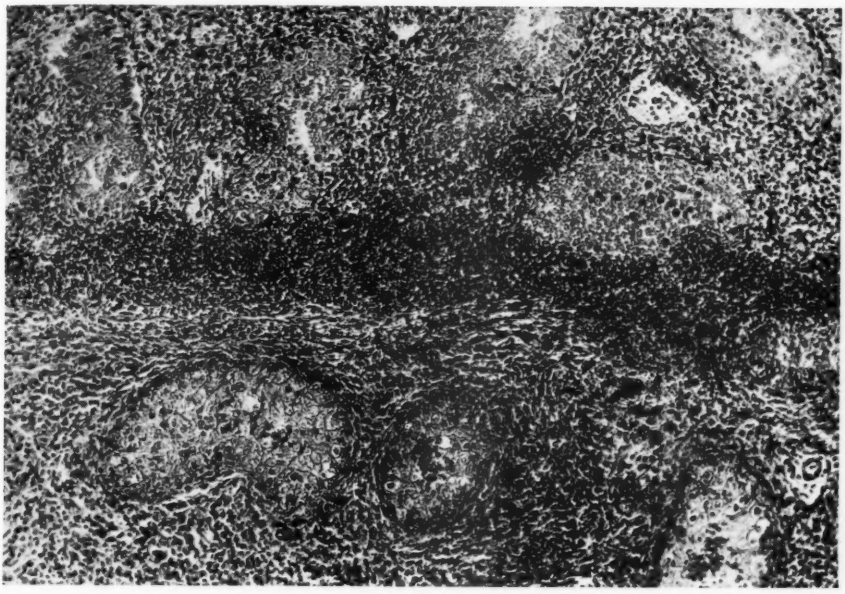

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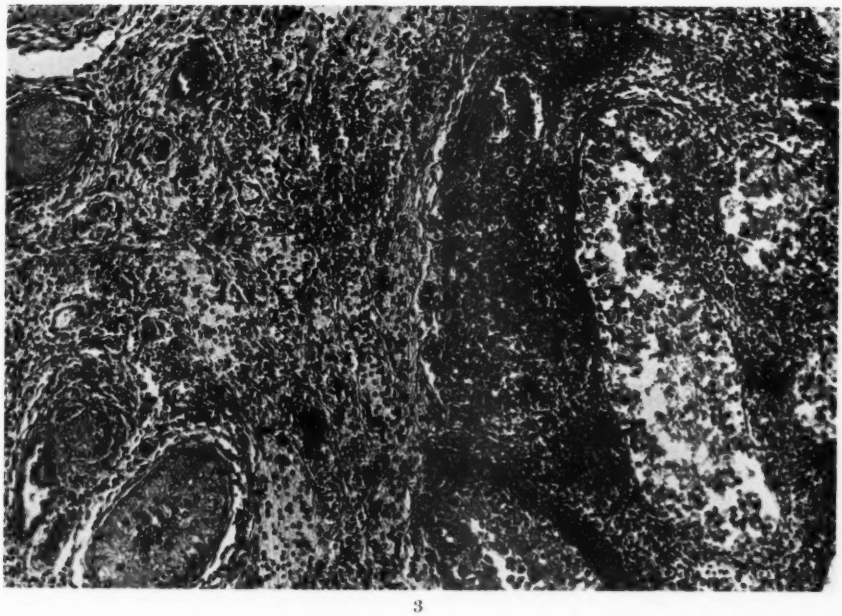

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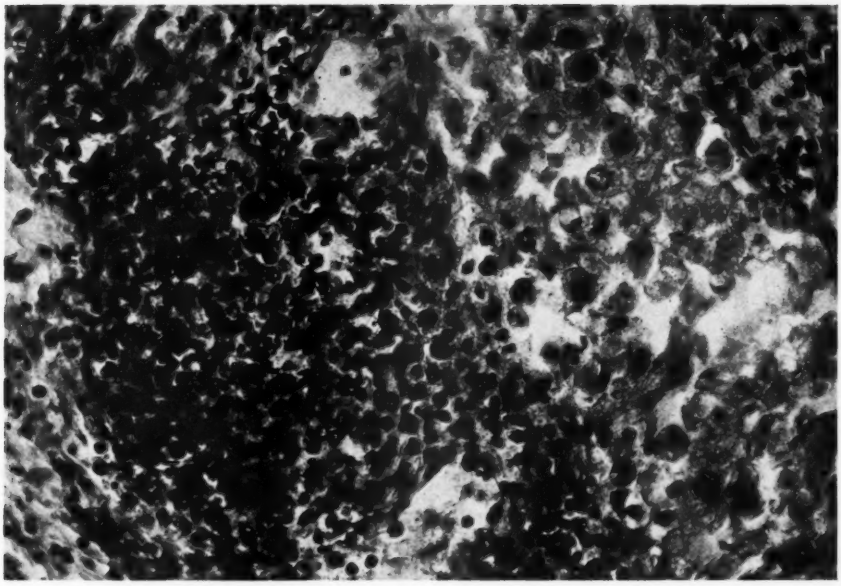

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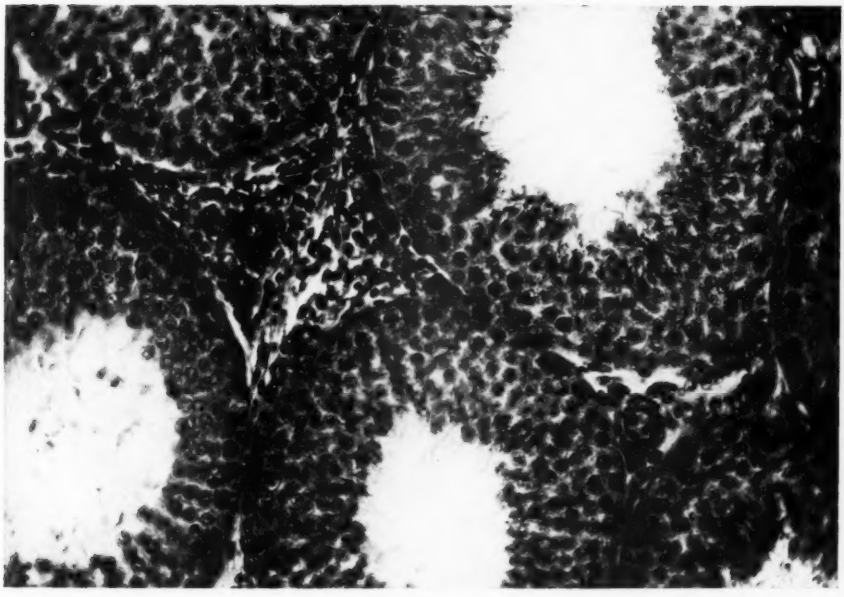

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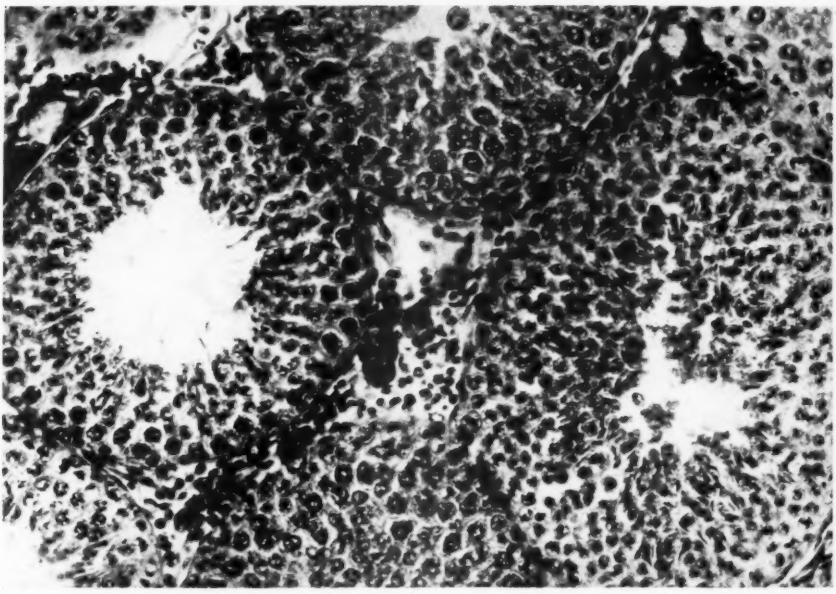
Stewart Focal Tuberculin Hypersensitiveness



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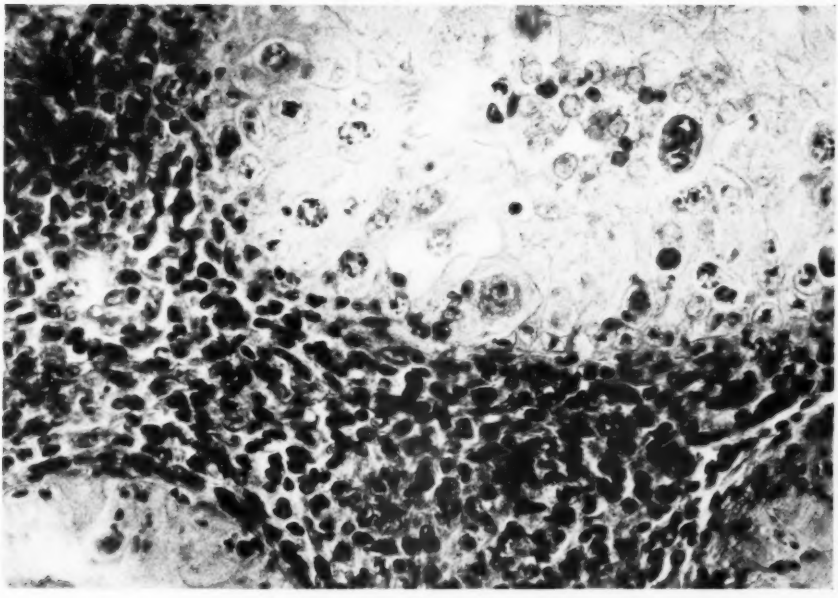

7

GROSS AND HISTOLOGIC CHANGES IN THE EDEMA OF PARAPHENYLENEDIAMINE *

M. L. TAINTER AND E. M. HALL

(From the Departments of Pharmacology and of Pathology, School of Medicine, Stanford University, San Francisco)

The striking edema of the head and neck regions appearing in about one and one-half hours after administration of paraphenylenediamine to rabbits involves mainly the tongue, conjunctiva, skin of the face and lips, submandibular tissues, larynx, and vocal cords. Anatomically, therefore, its location appears to be rather specific. Physiologically, the mechanism of its formation is concerned essentially with a marked increase in vascular permeability.[1] The response of cats to paraphenylenediamine [2] is similar to that of rabbits, except that edema of the larynx and vocal cords occurs more markedly and rapidly in cats than in rabbits. Studies of this experimental edema may ultimately have fundamental and practical bearings, especially in view of the occurrence of specific edema processes [3] in other regions from another compound (metaphenylenediamine) differing from paraphenylenediamine by the position of an amino group only.

As far as we know, histologic studies of the interesting edema of paraphenylenediamine have not been made previously. It is, therefore, the object of this paper to report the changes observed in ten rabbits with the desire of completing a study of various phases of the phenomenon by one of us (M. L. T.), and of demonstrating that the edema is specifically localized. Before discussing the histologic changes, a short summary of the gross changes at necropsy will be presented. This has been made from a careful study of the changes in over 350 rabbits and cats by one of us (M. L.T.).

CHANGES AT NECROPSY

At necropsy there was a variable amount of chemosis in the conjunctivae without hyperemia or signs of irritation. The skin of the face was usually edematous, the nose being about twice its normal

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width. The tongue was always markedly swollen, and on section appeared soggy and gelatinous. Frequently the tissues of the floor of the mouth were raised into large gelatinous bullae. In the neck, the edema was confined mainly to the area between the rami of the mandible, which was filled with a large collection of semi-gelatinous and translucent material. The gelatinous consistency of this material was due to the presence of fibrin, since it was previously demonstrated [1] that whole plasma with its fibrinogen escaped into the area of edema. The vocal cords were often in tight apposition because of the bullous swelling present, which at times extended to the mucosa and muscles of the larynx. The muscles of the head and neck frequently appeared blackened, as if stained by the drug or some changed form of it. This discoloration was never observed in other muscles of the body, except at the site of the injection where the paraphenylenediamine came into direct contact with the underlying musculature. The selective staining of the head and neck muscles may be regarded as further evidence of selective action of paraphenylenediamine. It was in general agreement with the selective escape of certain dyes in the same region reported in a previous paper.[1] The thoracic and abdominal viscera showed no characteristic gross changes except for variable degrees of passive congestion. There was no excess free fluid present in the pleural and abdominal cavities. Careful necropsies in over 350 rabbits and cats failed to reveal edema of other regions of the body, including the subcutaneous tissues, except at the site of injection where there was a variable amount of edema due to local irritation from the compound itself, and from the acidity of the hydrochloride. Histologic examination of a rabbit that had marked edema of the head and neck showed no edema at the root of the tail. Accordingly, therefore, we were not able to confirm Gibbs's claim[2] that edema produced by paraphenylenediamine occurs also in the paws and at the root of the tail in both rabbits and cats. The results of our gross studies justify the conclusion that paraphenylenediamine produces edema of the head and neck regions only. This is sustained by our study of histologic changes associated with the process, now to be described.

Histologic Changes

Methods. The tissues studied histologically were obtained at necropsy from ten rabbits used in connection with another study,[4] the details of which will be reported later. All the rabbits received subcutaneous injections of 0.2 gm. of purified paraphenylenediamine hydrochloride per kilo, dissolved in about 10 c.c. of water. In addition to the paraphenylenediamine, six of the rabbits received other treatments. Rabbit 219 received 11.6 mg. per kilo of nicotine subcutaneously in seven divided doses during the three hours of the experiment, and Rabbit 262, an infiltration of the left Gasserian ganglion with nicotine. In Rabbit 233 the left trigeminal nerve was cut in the skull with a Magendie neurotome ten minutes before the administration of the paraphenylenediamine. In Rabbit 247, the sensory nerves of the tongue, and in Rabbits 245 and 246, those of the tongue and neck, were cut and allowed to degenerate for about two weeks before the paraphenylenediamine was injected. These various procedures have been found [4] not to influence the usual course of the edema process. In addition to these rabbits, the tissues of two untreated rabbits were studied as controls.

Blocks of tissue removed at necropsy were placed immediately into Orth's solution for fixation. These were later embedded in paraffin. The sections were stained throughout with both hematoxylin–Van Gieson and hematoxylin–eosin solutions. The histologic changes in important organs of all rabbits studied are summarized in the accompanying Table. Fig. 1 illustrates a section of the tongue of an untreated rabbit for comparison with that of a typical edematous tongue after paraphenylenediamine illustrated in Fig. 2. In order to give some quantitative idea of the edema in the various organs and in different animals the following designations and their signs were adopted: Slight (+), for just recognizable edema; moderate (++), marked (+++), and extreme, (++++).

Head and Neck. The presence of edema in these regions was readily recognized by the wide separation of the tissue cells and cell-groups from one another, and by the presence of fibrin in the spaces. The quantity of fibrin present was often marked. Fig. 2 illustrates the marked separation of muscle fibers frequently observed in the tongue. The results in the Table leave no doubt that the most pronounced edema occurred in the tongue, larynx and

TABLE

Degrees of Edema Following the Subcutaneous Injection of Paraphenylenediamine Hydrochloride in Rabbits (0.2 gm. per kilo) (1)

Number of rabbit	Tongue	Larynx	Neck tissues	Masseter muscle	Thymus gland	Submaxillary gland	Other tissues
261 (control; untreated)	−	−	−	−	−	−	
334 (control; untreated)	−	−	−	−	−	−	Pectoral and Thigh Muscles −
230	++++	++++	+++	+++		+	Pectoral and Thigh Muscles −
253	+++	++	++++	++	−	+	Pectoral Muscle −
219 (2)	+++	++++	++	+++	−	+++	Skin of Nose ++
262 (3)	++++	++++	++++	+++	−	−	Pectoral Muscle −
233 (4)	++++	++	+++	+++	−	−	Pectoral and Thigh Muscles −
247 (5)	+++	+++	+++	Hyoid Muscle ++		++	Thigh Muscle −
245 (6)	++++	+++	+	++	+++		Thigh Muscle −
246 (6)	+++	+++	+++	+++	++	+	

1. In the table the negative sign (−) means no edema, and the positive sign (+) means edema was present with different degrees rated as follows: slight (+), moderate (++), marked (+++), and extreme (++++).
2. Nicotine 11.6 mg. per kilo subcutaneously in seven divided doses in addition to the paraphenylenediamine.
3. Left Gasserian ganglion infiltrated with 0.25 c.c. of 1 per cent nicotine in addition to the paraphenylenediamine, subcutaneously.
4. Left trigeminal nerve cut ten minutes before paraphenylenediamine.
5. Sensory nerves of tongue cut, and degenerated two weeks before paraphenylenediamine.
6. Sensory nerves of tongue and neck cut, and degenerated two weeks before paraphenylenediamine.

loose tissues of the neck. The masseter muscle showed almost as much edema as these organs. The thymus and salivary glands occasionally showed moderate degrees of edema.

Discoloration of Tissues. More or less discoloration was observed microscopically in the tissues in which the edema was most marked. In some cases the mucous glands of the larynx appeared almost black. The muscle fibers of the larynx were often discolored, and commonly the masseter fibers were very dark. The tongue and the muscles of the neck were also variably discolored. The dark coloration of these areas suggests some chemical action between the paraphenylenediamine and the tissues affected, perhaps an oxidase reaction. The significance of it, if any, for the specific edema process has not been investigated, though it may be that it is fundamentally concerned with vascular injury in the edema regions.

Gasserian Ganglia. Histologic examination of the Gasserian ganglia of Rabbits 245, 246 and 247, whose sensory branches of the trigeminal nerves were cut and degenerated two weeks before the injection of paraphenylenediamine, revealed rather marked chromatolysis involving approximately from one-third to one-half of the cells. Allen [5] has described similar changes in the Gasserian ganglia of cats following section and degeneration of some of the sensory branches. The Gasserian ganglia of Rabbit 349 with the nerves intact, and receiving paraphenylenediamine, showed no changes. Hence, it appears that the changes in the Gasserian ganglia of the denervated rabbits were not due to the paraphenylenediamine. Sections of the cerebral cortex, cerebellum, medulla and lumbar cord appeared normal except for moderate congestion. These sections did not differ in appearance from those of an untreated control (Rabbit 352).

Viscera. In Rabbit 245 the heart muscle showed moderate edema. In Rabbits 219 and 230 there was moderate edema of the lungs, which was perhaps due to, or accompanied death from, circulatory and respiratory failure.

The *liver* in most of the rabbits showed evidence of abundant glycogen content. This was further demonstrated in two rabbits by a special stain (Best's carmine). In three rabbits the liver was the seat of a chronic inflammation, and to this group belonged one of the controls (Rabbit 261). Rabbit 247 showed marked proliferation of the bile ducts and surrounding connective tissue, probably

the result of infestation with *Coccidium oviforme*, although none of the parasites could be found in the sections.

The *kidneys* presented granular swelling of the tubular epithelium in four animals, one of these being a control (Rabbit 261). Two of these were rabbits in which there was chronic inflammation of the liver. The kidney changes in the three rabbits showing edema were probably not due to the paraphenylenediamine, since it had been shown previously by Tainter and Hanzlik [1] that renal functional efficiency during the edema was not impaired and the kidneys of their five rabbits showed no pathological changes.

The *pancreas* showed moderate edema in Rabbit 246 only. It was normal in all the other rabbits.

The *spleens* of all rabbits throughout were normal except for varying degrees of passive congestion.

Skin. We studied the changes in the skin after applications of 5 per cent aqueous and alcoholic solutions of the base and the hydrochloride daily for three weeks to areas of about 2 cm. in diameter of the shaved abdominal skin of a rabbit. The areas to which the base was applied appeared moderately thickened at necropsy, this change being greatest in the area receiving the alcoholic solution of the base, and was limited to the areas of application. However, no hyperemia or edema could be seen. The areas receiving the hydrochloride showed no differences from the untreated control areas.

Histologically, the treated areas of the skin showed extreme edema which was not confined to the dermis, but involved the underlying muscle to a moderate degree. The dermis was fully three times its normal thickness. The collagen fibrils in it were widely separated and a fairly large amount of fibrin was present in the spaces. The epidermis, which was very thin, appeared unchanged or normal. Discoloration and congestion were absent in the tissue sections. Further experiments were not made as the form in which paraphenylenediamine is present in various cosmetic preparations, etc., is not known or is uncertain. Therefore, evidence bearing on dermatitis from such preparations could not be satisfactorily studied. The presence of other constituents in these mixtures may easily modify the skin response, although the repeated application of free paraphenylenediamine base undoubtedly can cause considerable injury.

Conclusions

1. The systemic administration of paraphenylenediamine hydrochloride to rabbits and cats produced a marked edema in the tissues of the head and neck.

2. Gross and microscopic studies of various organs and tissues in rabbits showed that the edema process was not general, but was sharply localized in the head and neck regions, and, therefore, rather specific.

3. Histologically, the edema was characterized by a wide separation of the tissue elements and an extensive precipitation of fibrin from the exuded lymph. This ready coagulation of the fluid indicates a severe type of injury to the blood vessels, presumably in the capillaries.

4. This was further sustained by the presence of darkening (discoloration) in certain edema tissues from the poison, this phenomenon agreeing with the selective escape of dyes in these regions previously reported.

5. Repeated local applications of alcoholic and aqueous solutions of paraphenylenediamine base to the skin of a rabbit produced marked edema at the site of application.

We wish to express our thanks to Doctors Ophüls and Hanzlik for suggestions and criticisms of the manuscript.

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DESCRIPTION OF PLATE LXXXVI

Fig. 1. Section of tongue of untreated Rabbit 334 showing normal appearance of epithelium and muscle. (Low power.)

Fig. 2. Section of tongue (same region as in Fig. 1) of Rabbit 262 which received 0.2 gm. per kilo of paraphenylenediamine hydrochloride subcutaneously. Shows hydropic changes in the epithelium and extreme edema of the muscle with abundant fibrin. (Low power.)

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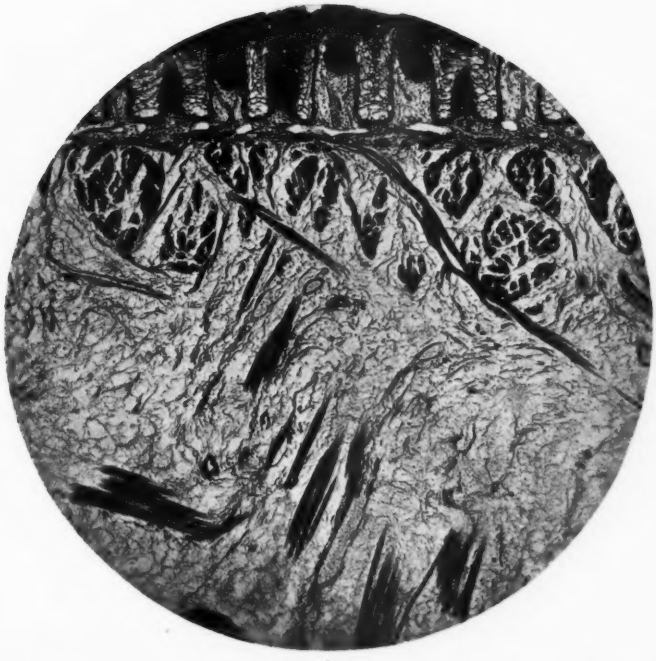

2

Tainter and Hall Edema of Paraphenylenediamine

MECHANICAL IRRITATION AS ETIOLOGIC FACTOR OF CANCER *

CLINICAL OBSERVATION

NICHOLAS M. ALTER, M.D.

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Virchow's[1] "Irritation Theory," promulgated in 1855, has remained the most plausible and fruitful hypothesis for the etiology of neoplasms. This theory states briefly that: "Irritation is the essential factor of neoplastic tissue proliferation." His general principles concerning neoplasms, concisely formulated as "*omnis cellula e cellula*" and many other of his principles have stimulated the study of tumors by persistent empirical-clinical observations and were largely responsible for the numerous other theories explaining the origin of tumors (Cohnheim, Ribbert, etc.). These clinical and morphological studies, in turn, have led to extensive and systematic experimental work which in the hands of such investigators as Fibiger, Rhodenburg, Yamagiwa, Ichikawa, Bloch and others, of recent years, has brought out startling and convincing evidence in favor of the theory.

One of the earliest general principles laid down by Virchow declares that "For the production of vital activity of any part of the body, excitation or, in other words, irritation is necessary." Applying this principle to the formation of neoplasms, he made the deduction that "The production of large groups of cells from single ones occurs in the adult body unquestionably as the result of direct irritation of the tissues."

By means of analysis of a rich collection of empirical observations and experimental data, Virchow developed further details of the irritation theory. He recognized numerous single factors, as for example: "In case of mechanical irritation caused by a thread, the swelling is due to division of cells." "If the skin becomes irritated in consequence of continued friction and the irritation is increased to a certain point, the epithelium will thicken and, if the proliferation is very energetic, it may lead to tolerably large tumor-like

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formations." "The same effect, cell proliferation through cell division, may also be produced by chemical irritation as by the application of a caustic substance." As definite clinical examples of such irritations, Virchow referred to lip cancers of pipe smokers, and to cancer of chimney sweepers. It was through the study of the latter that the importance of tar as a chemical irritant has been demonstrated, and that the isolation of a chemical compound from tar by Bloch led to the production of cancer in animals almost at will.

Besides the action of chemical, mechanical and thermic irritants (*causa occasionalis*), emphasis is laid upon the importance of the local reaction of tissues with subsequent thorough and complex changes comparable to those occurring in the fertilization of the ovum after the action of the spermatocyte. This tissue reaction forms the second important factor in tumor formation, the *causa praedisponens*. In a long series of diverse conditions of local pre-disposition, Virchow emphasized the importance of *locus minoris resistantiae;* for instance, where derivatives of ectoderm and entoderm meet (orifices of the body, lip, anus, etc.), or in the viscera, the narrowed places (flexures, etc.) of the intestines which, because of their positions, are more exposed to irritation. The hereditary element of the local predisposition, is demonstrated in the congenital naevi (*n. verrucosus*, *n. pigmentosus*, etc.). On the other hand, as examples of acquired predisposition, Virchow [2] brings up a series of post-inflammatory changes: scars, polypi, etc., and emphasizes mainly the general fibrosis following chronic inflammation. The mucous membranes deserve mention particularly on account of the frequent hyperplasia, incited by the chronic inflammatory irritation, which in turn may often lead to real tumor formation (polypi).

The third important factor in production of neoplasms is the general *dyscrasia*, which however has to be strictly distinguished from *constitutional disposition*. This latter involves the changes of the tissue juices and blood as well as the alteration of all the body tissues proper. On the other hand, *dyscrasia* according to Virchow is always secondary to some local tissue changes. The essential changes of *old age* he found in various tissues. These are followed secondarily by *dyscrasia*. Various single factors usually combine in the production of tumors; for instance such is the case when an undescended testicle (developmental defect) is exposed to trauma, a

combination of local misplacement and mechanical irritation. The various irritants, as single factors, usually combine for the production of neoplasms; according to Virchow, "inflammation never admits of a single explanation; we can find side by side all the forms of irritation."

This tumor theory opened a wide experimental field for cancer research which however has only recently made rapid progress. For such work accurate clinical and morphological observations form a firm working basis. The purpose of this paper is to record a singularly interesting clinical observation in which the mechanical factor seems to be predominant in the production of cancer. The subsidiary factors must necessarily play a significant rôle, but the degree of their importance is obscure.

Case Report

Patient W. L. K., male, 60 years old, white, was admitted to the service of Dr. K. I. Sanes in the Western Pennsylvania Hospital, Pittsburgh, Pa., on Aug. 15, 1915, with the chief complaints of (1) abdominal pain, (2) constipation, (3) loss of weight, (4) anorexia.

Family History. Mother strikingly emaciated, died of old age.

Past History. Negative.

Present Illness. Began about one year before admission to the hospital with obstinate constipation. Six months later patient began to suffer from severe abdominal pain, which was pelvic in site and which often awakened the patient from his sleep. Later these attacks of pain with the same localization occurred during or immediately after meals. About two weeks before admission to the hospital patient had very severe attacks of griping pain of the same type as mentioned before, with nausea but without vomiting. His constipation became very bad and patient began gradually to develop obstructive symptoms. He lost 36 pounds in weight within the past six months (from 171 to 136 pounds).

Physical examination of the abdominal cavity revealed a moveable hard mass, probably of the sigmoid. Blood count was negative. Urine contained some acetone. Stool examination showed the presence of blood. Roentgenologic examination showed complete obstruction of the sigmoid somewhat below the end of the descending colon. Bismuth meal did not pass into the sigmoid. The bismuth column showed the same position at the end of thirty-eight hours. The bismuth enema passed this place although it did not fill up the descending colon.

Clinical Diagnosis. Carcinoma of the sigmoid.

Operation of sigmoidectomy was performed by Dr. K. I. Sanes at the Western Pennsylvania Hospital on August 19, 1915. A hard, constricting growth of the sigmoid was found below the junction with the descending colon. The growth had not invaded the serosa which had a smooth surface, but with some cicatrization of the meso-colon involving the inferior mesenteric artery. About 12 inches of the large intestine were resected with electric cautery. The growth occupied about the middle of the removal bowel. The operation was compli-

cated by interference with the blood supply of the lower bowel. The inferior mesenteric artery had to be ligated, as it was involved in the above-mentioned cicatrix. Consequently the descending colon was removed for the most part and the remnants of the colon and the sigmoid were united in an end to end anastomosis. The abdominal cavity showed nothing unusual otherwise. The liver was slightly enlarged, but no hard nodules were discernible in it.

The patient's convalescence was uninterrupted and after complete recovery he was discharged from the hospital on September 13, 1915. The last roentgenologic examination, in March, 1922, revealed the formation of a slight diverticulum at the place of the anastomosis. The patient, however, had made no complaints; his bowel movements and digestion have given him no trouble and he is still enjoying good health at present.

Pathologic Report

Gross Examination. Specimen consists of a piece of large intestine 30 cm. in length (Fig. 1). At about its middle portion a circular constriction is seen, which is hard in consistence and is covered with scar-like depressions and fibrous tags, but nothing else is noticed on the outside. Above the constriction the bowel is obviously distended, its wall is leathery and thickened as compared with the portion below the constriction. On opening, the knife meets with increased resistance when cutting through the constriction. The lumen shows a striking picture. There is an annular, ring-formed elevation constricting the lumen. It measures 4.5 cm. in width and averages 2.5 cm. in thickness. On cut surface a gray growth is seen with bacon-like, transparent appearance which shows gradual transition at its base to tendinous fibrous tissue, and at its periphery to yellow, subserous fatty, material. This ring-formed constriction has rather sharp, elevated edges and a depressed, crater-like middle portion. The surface of the crater is covered with hemorrhagic and necrotic material. The lumen here measures about 1.5 cm. in diameter and is filled completely by a round polyp. This has a mulberry surface covered with mucus and is soft in consistence. It forms the head of a long pedunculated growth with a broad attachment 8 cm. above. The stem is fan-shaped at its attachment (8 mm. at its base) and tapers to a neck where it measures 3 mm. in thickness. The round head of the polyp has a valve-like action and can be dislodged from below. It fits in snugly in the crater-like center of the annular growth, as a ball in a socket. On cross section, the head of the polyp shows soft consistence and rich mucoid secretion of the surface layer. A peach stone is lodged above the entrance of the constriction and

presses against the polyp. The stone is slightly larger than the lumen at the constriction and has otherwise the usual, rather rough surface and edges.

Above the constriction the mucosa is somewhat thickened and covered with mucus. The musculature shows obvious hypertrophy. The muscle layer measures 4 mm. in places. On one side there is a peculiar, rather superficial pouch lined with mucosa as elsewhere but with strongly developed musculature. The pouch seems to have been the nest for the peach stone.

Microscopic Examination. Section taken from the annular constriction at the crater-like ulceration shows a malignant epithelial growth. The glandular structure of the growth is well preserved in most places. The transition from the normal epithelial covering to the irregular epithelial proliferation can be followed distinctly (Fig. 2). The malignant cells vary a great deal in size and shape. In the glandular portions they are rather cylindrical with irregularly placed nuclei which show numerous mitotic figures. In the scirrhous portion, the cells are quite small. In the central portions the surface is necrotic. In the deeper layers the cells are smaller and surrounded by large amounts of dense fibrous tissue. This scirrhous growth, however, has not invaded the subserous tissues. The stroma increases in amount towards the deeper layers and contains a marked infiltration of lymphocytes. Section of the head of the polyp shows a benign epithelial tumor (Fig. 3). The surface shows typical intestinal glands, mostly perpendicular to the surface. In the deeper layers the glands run an irregular course and consequently they are cut in various planes. The cells are very regular in every respect. They are cylindrical with nuclei placed near the basal membrane. The blood vessels are well formed. Around the hilum they have also a muscular wall and are surrounded by fibrous stroma which contains scant lymphocytic infiltration. The surface epithelium consists mainly of goblet cells with increased mucus secretion which also covers the surface of the growth. Sections of the stem and base of the polyp show dense fibrosis and rich lymphocytic reaction. There are quite a few eosinophilic leukocytes present. Some sections of the intestinal wall above the constriction show marked hyperplasia of the smooth muscle cells. Van Gieson's and Mallory's connective tissue stains reveal an increase of fibrous tissue with diffuse lymphocytic infiltration.

Anatomic Diagnoses. Scirrhous carcinoma of large intestine; pedunculated adenoma of large intestine; dilatation of intestine and muscular hypertrophy of intestinal wall above constriction.

Discussion

From the pathologic study of the specimen it is evident that we are dealing with a carcinoma of the sigmoid, which originated where the head of the polyp caused friction of long duration. Obstruction was brought about by the ball valve-like mechanism of the polyp and cancerous constriction. Above this there is a marked muscular hypertrophy of the intestinal wall. Primarily this hypertrophy was due to the effort of the intestine to expel the polyp. This effort of expulsion pulled out the polyp and produced an unusually long stem (8 cm.). Unquestionably a long time was necessary to accomplish this. Later on, when a constriction was brought about and thus prevented the progress of the peach stone, the attempt to expel the stone produced an additional muscular hypertrophy by the same mechanism, with the result of a pouch-like irregularity of the intestinal wall. These conditions seem to indicate a well-marked mechanism of long duration. The formation of the polyp was probably due to a previous chronic inflammatory process. There is strong evidence for the causative relation between the head of the polyp and the cancerous growth, which corresponds accurately to each other as to location where the friction occurred, as a ball in a socket. This is particularly striking in view of the long stem. All of this seems to exclude a mere coincidence. The peristalsis of the intestines produced the continuous to-and-fro friction which had been increased by the effort of the intestines to expel the polyp and the peach stone. The head of the polyp, perhaps, did not remain in the place of constriction for a long period. This may explain the clinical observation that the patient never had complete obstruction. Still another factor of mechanical irritation may be added in the nature of fecal concretions which seem to play an important rôle in the production of cancer at the various flexures of the large intestine.

There are numerous empirical observations as to neoplasm caused by chronic mechanical irritation. Bashford[3] reported cases of melanosarcoma occurring in natives of Africa, which are caused by thorns causing continuous irritation of the sole of the foot. Cancers caused

by irritating foreign bodies (pessaries, etc.) have often been reported. Very striking also is the "cancer of the horn core" of Indian cattle, which develops at the root of the right horn used for attachment of agricultural implements. No cancer develops on the left horn of these cattle. Stones in gallbladder and renal pelvis as well as cancers of the tongue, which correspond to bad teeth and lip cancers of pipe smokers, have an inflammatory element in etiology besides the pure mechanical irritation. In the case reported in this paper, both the cancer and the polyp have the inflammatory factor in their etiology. This is unquestionably of primary importance particularly as far as the formation of the polyp is concerned. The proof of this is seen in diffuse fibrosis and lymphocytic infiltration at the base of the polyp. Post-inflammatory fibrosis has been recently emphasized in the so-called precancerous-lesions. The older and ancient authors used the term of "scirrhous induration" not only for cancers, but also for inflammatory fibrotic lesions. Galen[4] stated that *polypi aut inflammatione aut tuberculo*. . . . Galen emphasized the importance of inflammatory lesions and "other nodules" preceding the formation of cancers besides the general dyscrasia, *atra bilis*.

This has been given a strange, literal interpretation, whereas it represents the humoral pathologic conception of Hippocrates and Galen. It has been accepted also by Virchow, although he considered the humoral changes secondary to the tissue changes as seen in old age. Local cancerous predisposition of the sigmoid can be assumed on an embryological basis, also shown by the peculiar development of the mesosigmoid as contrasted with the mesocolon above and below it.

Conclusions

1. The case of carcinoma of intestine reported in this paper appears to present a striking example of mechanical irritation as an etiological factor in the histogenesis of cancer.

2. The case is analyzed and best understood by means of Virchow's "Irritation theory." According to this any of the single factors of three groups may be predominant in the histogenesis of cancer, which, however, is usually the result of various combinations of them.

These groups are, (*a*) *Causa occasionalis*, as the various irritants: mechanical, chemical, or thermic.

(*b*) *Causa predisponens*, as the embryological anlage or post-inflammatory changes.

(*c*) *Dyscrasia*, as general constitutional change.

The various well-known cancer theories and recent experimental works (Conheim's, Ribbert's tar cancers, etc.) represent only emphasis and elaboration of single factors in Virchow's comprehensive theory.

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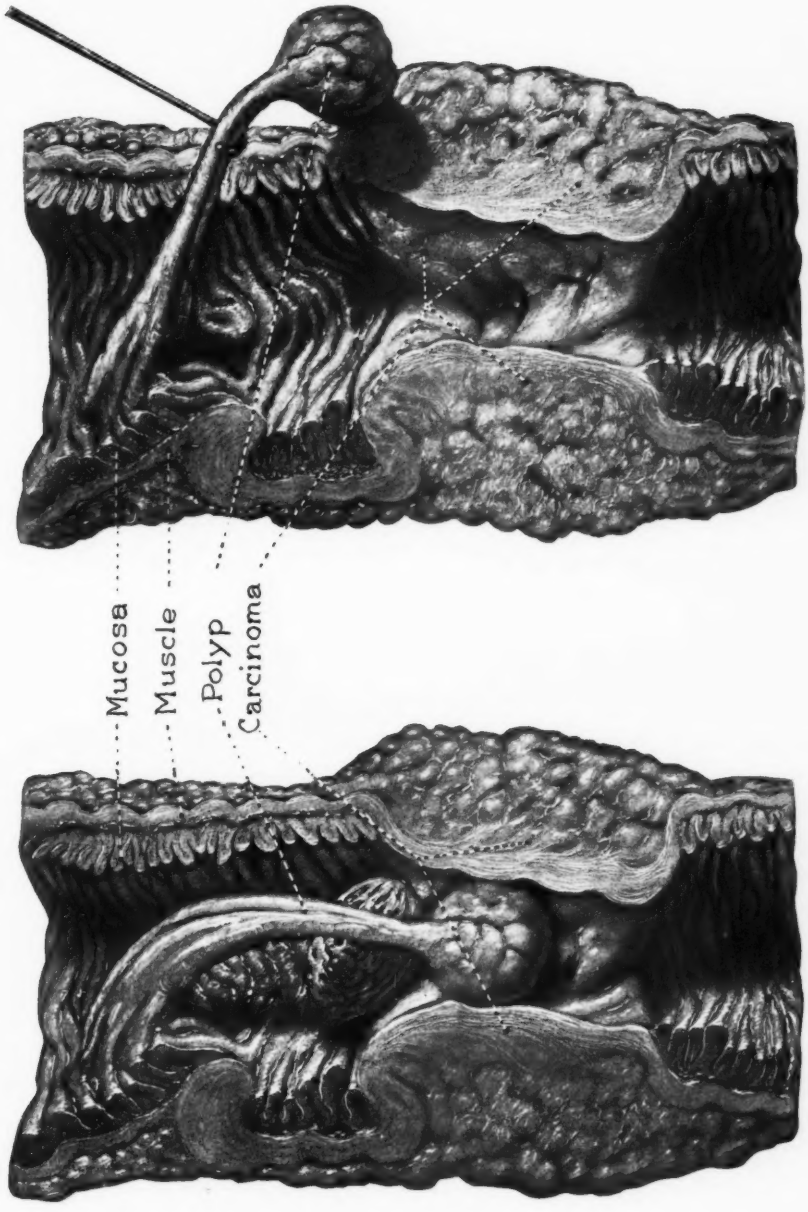
DESCRIPTION OF PLATES LXXXVII–LXXXVIII

Fig. 1. Drawing of specimen, showing lumen of intestine. *At left:* Annular constriction of scirrhous carcinoma. Lumen is completely plugged by head of polyp with long stem (ball in socket). Above head of polyp is the peach stone. *At right:* Polyp is lifted out of its nest. Peach stone is removed.

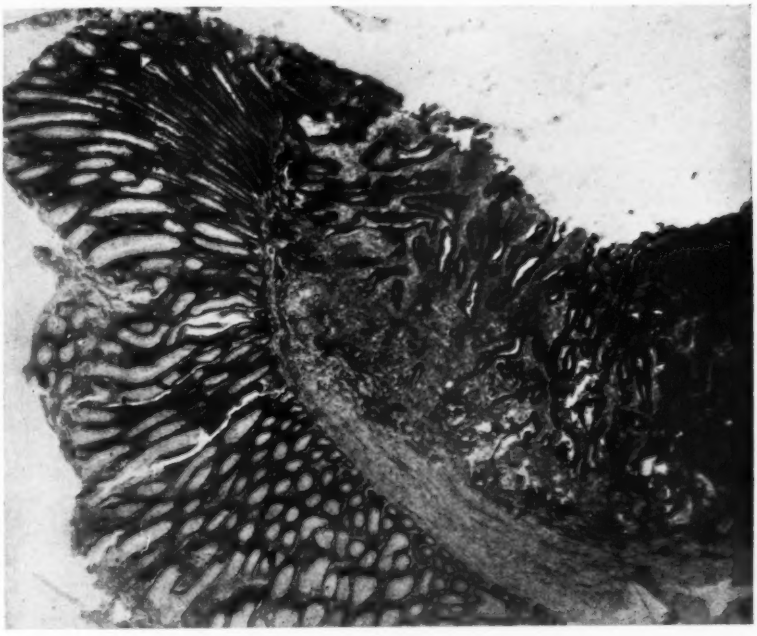
Fig. 2. Photomicrograph of section of intestinal wall, showing transition of normal epithelium to malignant proliferation. X 35.

Fig. 3. Photomicrograph of section of head of polyp, showing intestinal adenoma. X 35.



Alter

Mechanical Irritation



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Alter Mechanical Irritation

NEUROBLASTOMA OF THE INTESTINE *

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Embryologists agree that the primitive undifferentiated nerve cells are of ectodermal origin, and early in the course of the histogenesis of the nervous system these cells differentiate into spongioblasts and neuroblasts. The former develop the supporting neuroglia structures of the central nervous system, while the latter are the precursors of neurons and also of the migratory cells which spring from the ganglionic crest, the anlage of the sympathetic nervous system. Some of these neuroblasts migrate ventrally toward the aorta to form the sympathetic chains with their paraganglionic structures and also invade the mesodermal part of the suprarenal glands to form their medullae. It is believed by some (Kohn [1]) that such primitive nerve cells may come to rest anywhere in the body, remain in a latent state and at some later time may proliferate and become neoplastic.

Marchand, in 1891,[2] reported a tumor of a suprarenal gland in a child 9 years old, which was rich in cells and fibrils, resembling the tumor now recognized as neuroblastoma. The presence of the fibrils led him to consider it a glioma.

Küster, in 1905,[3] reported two similar cases; one, a child 3 months old, presented a primary tumor of the right suprarenal gland with metastases in the liver; and a second case in which the tumor was also primary in the suprarenal gland, not complicated by metastases. He described undifferentiated nerve cells associated with fibrils, the latter staining brown with Van Gieson's method. He was the first to draw attention to the occurrence of a peculiar cell-grouping, the so-called "rosette" formations. He considered these tumors malignant gliomata.

Wiesel,[4] in the same year, called attention to the similarity between the cellular picture of the tumors described by Küster, and that of primitive sympathetic structures and of the medulla of the suprarenal gland.

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In 1910, J. G. Wright [5] reported four similar cases and definitely described the neoplasm as a neuroblastoma. He was able to demonstrate a very close similarity in its structure to the primitive sympathetic nervous system and the medulla of the suprarenal gland. This work was later confirmed by Landau.[6]

The latter reported three cases, and stressed the fact that the fibrils need not necessarily be present in these tumors. He believed that the more malignant the tumor, the fewer the fibrils and the more cellular its structure; and suggested that the younger the host, the more embryonic in character and more malignant is the tumor. He also showed that ganglioneuroma can be regarded as merely a more mature and more highly-differentiated form of neuroblastoma.

Pick and Bielschowsky [7] conversely pointed out that malignant neuroblastoma is the unripe counterpart of ganglioneuroma and therefore named these tumors *ganglioma embryonale sympatheticum*.

Pick, in 1912 [8] brought the number of cases up to eighteen, including one of his own. He discussed the histogenesis and relation of this form of tumor to the primitive nerve cells, and drew attention to the fact that in maturing the cells differentiate into higher, more specialized forms, the ganglion and paraganglion cells. Further contributions to the subject of neuroblastoma with extensive reviews have been made by Wahl,[9] Harbitz,[10] Lehman,[11] and R. A. Lambert.[12] Wolbach and Morse, in 1918,[13] brought the total number of undoubted cases up to twenty-nine, including three of their own. They, however, did not include the two cases published by Harbitz. Since then, only a few cases of this relatively rare neoplasm have been reported, Gunby,[17] Anderson and Shennan,[18] and J. H. Berner.[19]

The sites of primary occurrence in the reported cases were the suprarenal glands, sympathetic ganglia, retroperitoneal tissues, coccygeal gland, nose and uterus.

The observations to date have indicated that the term neuroblastoma must be reserved for tumors composed only of the undifferentiated type of nerve cell. On the other hand, tumors composed of ganglion cells with well-defined nerve fibers are called ganglioneuroma; and, since they are highly differentiated, are usually benign, in contradistinction to neuroblastoma. The third form of neurogenic tumor is the paraganglioma which is composed largely of chromaffine cells.[21]

Where mature ganglion cells have been found in typical neuro-

blastomata, they are to be regarded as forms in which partial transition has taken place from the primitive neuroblast to the mature ganglion cell. (Pick,[8] Landau,[6] Martius,[14] Wahl,[9] and Dunn.[15])

Martius reported a rather unusual case in which the tumor presented two almost separate parts, one simulating neuroblastoma, the other a ganglioneuroma. Other authors have reported cases of ganglioneuroma in which there occurred inclusions or islands of neuroblastoma (Pick und Bielschowsky,[7] Freund,[16] Dunn,[15] and others). Mixed neurogenic tumors containing all three elements of ganglioneuroma, neuroblastoma and chromaffine cells have been reported by Wahl [9] and others.

The two cases of neuroblastoma which I wish to add to the series already reported are unique because of their location and origin. In both instances the tumor arose in the jejunum, presumably from the nerve cells of the intestinal sympathetic plexus (Meissner's and Auerbach's). A search of the literature has failed to reveal reports of the occurrence of neuroblastoma arising in the intestine either primarily or by extension. The relative rarity of neuroblastoma occurring in the adult is an additional feature of interest.

Report of Cases

Case 1. Patient H. A., a widow, 55 years old, was admitted to Dr. Brill's service at Mount Sinai Hospital, New York City, on March 1, 1924, complaining of increasing constipation for four months. She attributed her complaint to an accident which occurred in 1923, when she was struck by a taxicab and sustained an injury to her right hip. The constipation was obstinate and required drastic cathartics for relief. There were no abdominal pains nor cramps, no nausea, vomiting, jaundice or melena. Her stools were usually firm, formed and dark brown in color. She had lost about 30 pounds in the preceding four months. Her past history was essentially negative. She had been a widow for twenty years and was never pregnant. There was no history of operations or serious illnesses. She had passed the climacterium twelve years ago.

Physical examination on admission to the hospital showed her to be well developed but markedly emaciated. There was moderate bilateral exopthalmos. The pupils reacted sluggishly to light. The heart was slightly enlarged and a systolic murmur was audible over the apex. The abdomen was distended. On palpation a large, oval, smooth, hard and immovable mass could be felt in the epigastrium, extending downward to the level of the umbilicus, more to the right than to the left of the midline. There was marked generalized voluntary rigidity of the abdominal muscles. The genitalia, aside from marked senile atrophic changes were negative. Examinations of the stools gave repeated positive guaiac reactions. The blood count, blood Wassermann test, and blood chemistry were negative. The phenolphthalein test revealed excretion of 80 per cent of the dye in the urine in two hours. Roentgen ray examination of the

colon with the aid of a barium enema showed the sigmoid to be markedly compressed by an extrinsic intra-abdominal mass.

An exploratory laparotomy by Dr. J. Brettauer disclosed a large neoplastic mass, densely adherent to the anterior abdominal wall. On separating the adhesions as well as the adherent omentum from the upper aspect of the tumor, the latter was brought into full view. The mass was about 8 cm. in diameter, grayish in color and had a rough surface. It appeared to occupy the upper portion of the mesentery and in it were incorporated loops of the upper portion of the jejunum. The large intestine encircled the mass, but was not invaded by it.

Five days after the exploratory laparotomy, the patient died from an extensive postoperative bronchopneumonia.

Necropsy. The body was that of an emaciated woman 55 years of age. Rigor mortis was complete. No icterus or petechiae were present. A 15 cm. median, recent abdominal operative incision was present which had been closed with interrupted sutures. Upon opening the peritoneal cavity a large, oval, firm, grayish-white, neoplastic mass measuring 15 x 10 cm. in diameter was found, which involved several loops of jejunum 5 cm. distal to the fossa duodenojejunalis and the contiguous mesentery. There were many recently formed fibrous adhesions between the parietal peritoneum of the anterior abdominal wall, omentum and the anterior surface of the neoplasm. The tumor apparently arose in the wall of the jejunum and by direct extension involved the walls of several adjacent loops of the jejunum and extended backward, distorting and invading the mesentery and the mesenteric lymph nodes in this region for a distance of about 5 cm. from its main attachment. As the result of erosion of the walls of the adjacent loops of intestine by the invading neoplasm, an intercommunication was established among several loops of the jejunum so as to form a cavity, the wall of which was formed partly by intestine and partly by tumor. The anterior surface of the mass, after separation of fibrino-purulent omental adhesions, was found to be greenish-yellow in color and necrotic in places. The free surface of the cystic cavity of the neoplasm was thrown up into large, firm, salmon-pink colored, irregular excrescences, covered by a film of thick mucus. Here and there necrotic areas were found. Cut section of solid portions of the tumor presented a homogenous, pinkish-white, friable surface.

The duodenum and the jejunum proximal to the site of the neoplasm were widely dilated, due to partial constriction of the lumen of the jejunum at the site of the neoplasm. The distal portion of the jejunum and the ileum were collapsed. The large intestine encircled

the neoplasm without being involved in the growth. On the basis of gross observation a tentative diagnosis of sarcoma of the intestine was made (Figs. 1 and 2).

The lungs were the seat of an extensive postoperative bronchopneumonia. The suprarenal glands were normal.

Examinations of all other organs revealed no significant findings.

Microscopic anatomy. Microscopic study of the tumor shows it to be essentially composed of numerous small, round, "lymphoid" cells, containing deeply stained nuclei, rich in chromatin granules and surrounded with a very little cytoplasm. Scattered throughout the microscopic field, a fine fibrillar stroma is seen which stains brown with Van Gieson's method but does not give the characteristic neuroglia stain with Mallory's hematoxylin phosphotungstic acid method. The cells present an alveolar arrangement and here and there are polarized in double rows about a central mass of finely granular material, assuming characteristic "rosette" formations. Some of the cells are somewhat larger in size and show mitotic figures. An occasional giant cell can also be seen. A few pyriform cells are present whose cytoplasm extends outward in the form of delicate fibrillar processes. No mature ganglion cells can be found. Numerous thin-walled blood vessels are seen in the reticular network supporting the cellular elements (Figs 3 and 4).

The microscopic characteristics of this tumor led us to group it with the undifferentiated type of neuroblastoma. This diagnosis was concurred in by Dr. F. S. Mandlebaum and by Prof. Ludwig Aschoff who examined the sections during his recent visit to New York City.

CASE 2. Patient, J. F., white, male, Russian, 64 years old, was admitted to Mount Sinai Hospital on December 17, 1924, service of Dr. A. A. Berg. The patient had complained of severe epigastric pains and distress for several months with loss of weight and occasional vomiting. There was marked constipation. No hematemesis or melena had been observed.

On physical examination the patient was found to be markedly emaciated. His heart was slightly enlarged and the sounds were of poor quality. An indefinite resistance was felt in the left hypogastric region, but no distinct tumor mass could be palpated.

Roentgen ray examination of the gastrointestinal tract revealed a partial obstruction high up in the small intestine. The stomach was negative except for slight residue after six hours.

On December 12, 1924, an exploratory laparotomy was performed, under local anesthesia, by Dr. Berg. He found a neoplasm involving the jejunum commencing about 7.5 cm. from the fossa duodeno-jejunalis and extending dis-

tally in the wall of the intestine for a distance of 8 cm. There were metastases in the adjacent mesenteric lymph nodes. Many small metastatic nodules were present on the serous surface of the jejunum, just adjacent to the main tumor. Twenty-two centimeters of the jejunum were resected together with a portion of the mesentery, containing tumor and enlarged glands. An end to end anastomosis was then made in the usual manner. The omentum was sutured over the anastomosis with interrupted catgut sutures and the abdomen closed without drainage.

The patient made an uneventful recovery. His appetite increased and his bowels moved without catharsis. His general condition improved very rapidly and he was discharged from the hospital on July 18, 1924.

The macroscopic specimen consisted of a portion of jejunum which measured 22 cm. in length and 9 cm. in its widest diameter, the narrowest diameter being 4 cm. Attached to the serosal surface infiltrating the mesentery and wall of the intestine there was a cellular growth measuring 5 x 2.5 cm. This neoplasm was situated directly opposite the narrowest portion of the jejunum, namely the area which measured 4 cm. in diameter. It involved the wall of the intestine at this point and extended distally for a distance of 8 cm. The mucosa of the involved portion of intestine was irregular, nodular and firm. On the serosal surface there were several small, round or oval, firm, neoplastic elevations which on section were similar to the main neoplasm (Figs. 5 and 6).

Upon microscopic examination the tumor is found to be made up of small, round "lymphoid" cells whose nuclei stain deeply and contain chromatin granules. The stroma of the tumor is composed of a fine, irregular, diffuse, fibrillar network which does not give the characteristic neuroglia stain. Scattered throughout the microscopic field are many pyriform-shaped cells with a round base, and pointed apex which gives off a fine fibrillar process. Their deeply staining nuclei are oval in shape, vesicular in character and contain many chromatin granules. Many giant cells of variable sizes are also present, some of which show mitotic figures. They are not of the foreign body variety (Fig. 7).

An interesting point in the histological structure is its resemblance to spongioblastoma, a form of neoplasm derived from the primitive glial elements of the central nervous system described by Strauss and Globus.[20]

Summary

Two cases of intestinal neoplasma are reported. They are grouped with the neuroblastomata because of their histological characteristics.

In both instances the tumors arose in persons past middle age. Metastases were found only in the adjacent mesenteric lymph nodes. The relatively benign character, at least in one of the tumors (Case 2), may, perhaps, be explained by its histologic resemblance to a form of tumor designated as spongioblastoma.[20, 21]

The observations indicate a new and hitherto undescribed site for primary neuroblastomata. So far as can be ascertained, no similar cases have been reported in the literature.

These cases emphasize the necessity for careful histologic study of atypical tumors, which on first examination might appear to be sarcomatous in character. It is probable that for this reason other instances of neuroblastoma of the intestine have been classified as sarcomata.

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DESCRIPTION OF PLATES LXXXIX-XCI

Fig. 1. (Case 1.) Anterior view of intestinal tumor showing its origin and relations.

Fig. 2. (Case 1.) Undersurface of tumor showing also partial obstruction of the intestine.

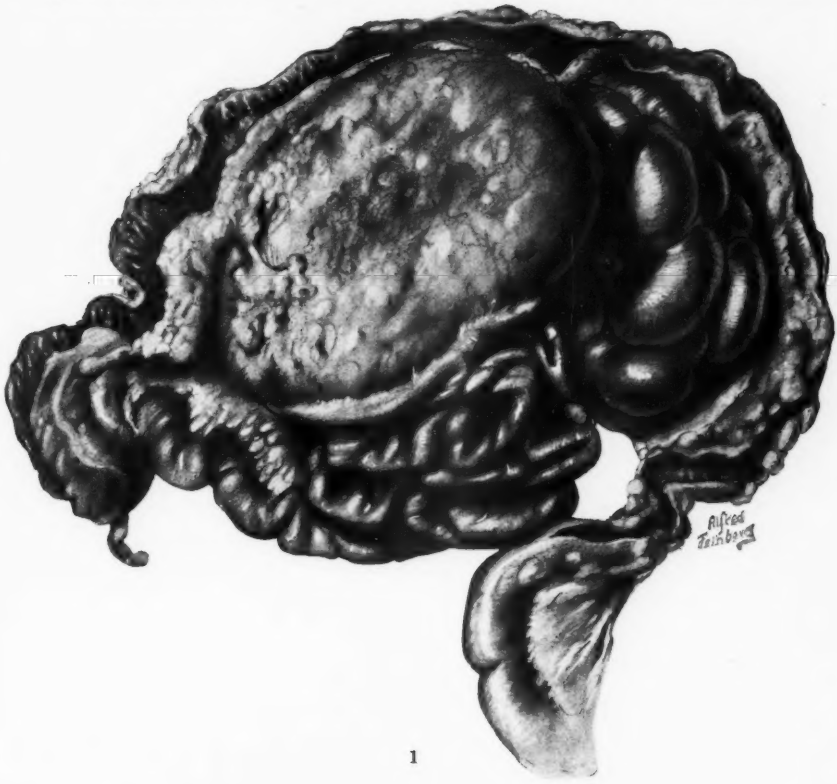
Fig. 3. (Case 1.) Photomicrograph of tumor showing "lymphoid" cells containing deeply stained nuclei surrounded by very little cytoplasm. Cells present an alveolar arrangement. A fine fibrillar stroma is scattered throughout the field. (High power.)

Fig. 4. (Case 1.) Section showing typical rosette arrangement of cells. (Higher magnification.)

Fig. 5. (Case 2.) Drawing of specimen of tumor of jejunum; view of mucosal surface.

Fig. 6. (Case 2.) Serous surface of tumor of jejunum with adjacent portion of invaded mesentery and lymph nodes.

Fig. 7. (Case 2.) Photomicrograph of section of tumor showing presence of large numbers of giant cells and numerous pyriform-shaped cells alternating with small "lymphoid" cells. For detailed description see text.



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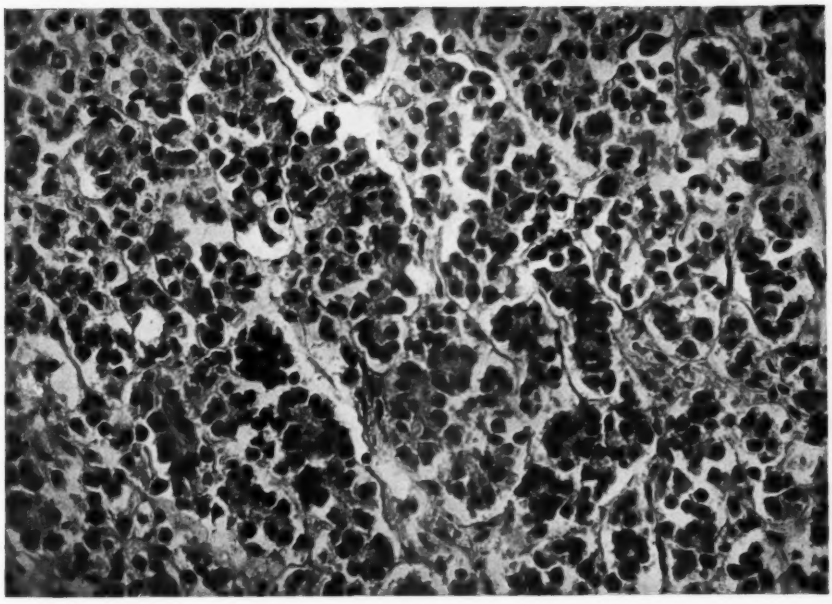
Ritter Neuroblastoma of the Intestine



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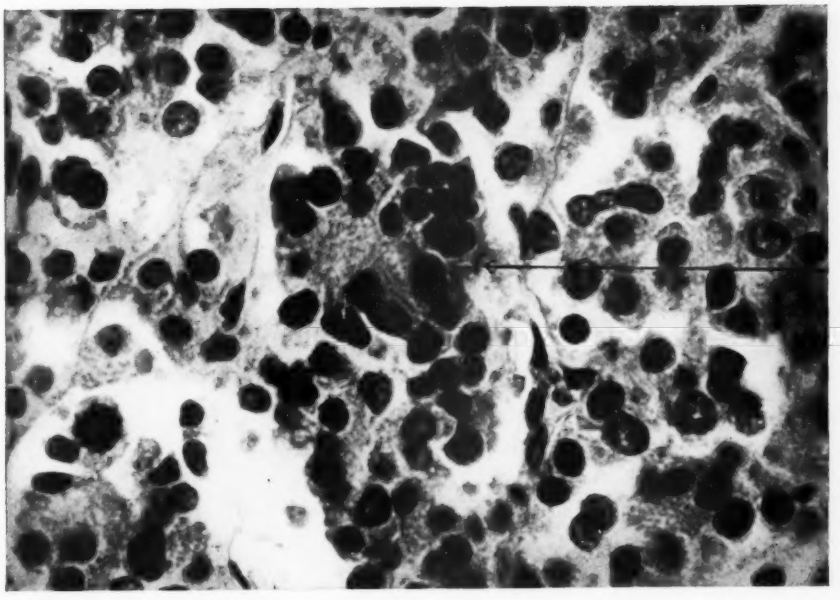

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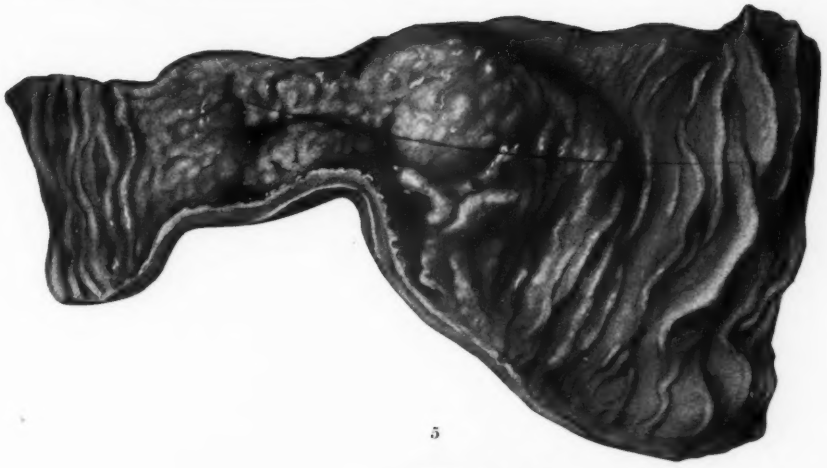

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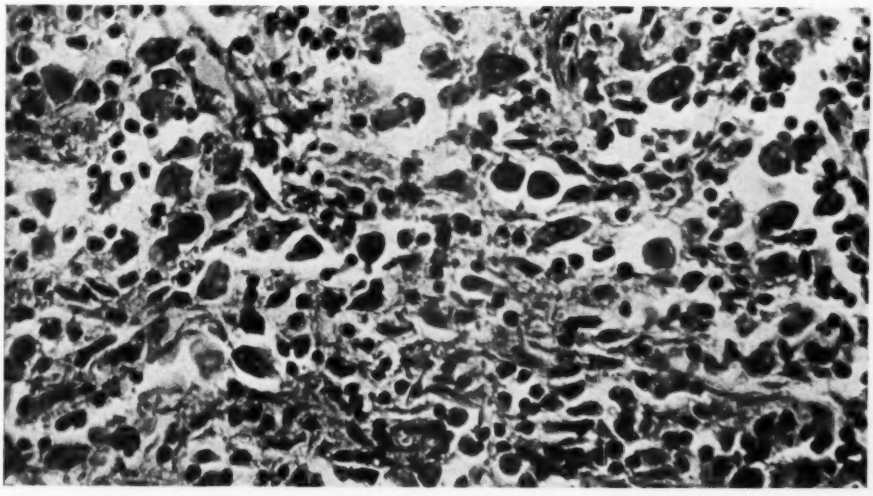

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Ritter Neuroblastoma of the Intestine

SCIENTIFIC PROCEEDINGS OF THE TWENTY-FIFTH ANNUAL MEETING

OF THE

AMERICAN ASSOCIATION OF PATHOLOGISTS AND BACTERIOLOGISTS

WASHINGTON, D. C.

MAY 5 AND 6, 1925

AMERICAN ASSOCIATION OF PATHOLOGISTS AND BACTERIOLOGISTS

SCIENTIFIC PROCEEDINGS

Owing to unavoidable circumstances it has been impossible to include the discussions.

THE VERNES FLOCCULATION TEST AS A GUIDE TO TREATMENT OF SYPHILIS.* A. B. Baylis, A. E. Sheplar, and W. J. MacNeal, New York.

Abstract. With adequate equipment the Vernes test on blood serum is less time-consuming than the Wassermann test and its results are recorded in precise figures to the second decimal place as milligrams of flocculation per cubic centimeter, through a range from −0.10 to +2.50. The Vernes reading has been aptly compared to the reading of a thermometer. It shows very slight fluctuations in health and in diseases other than syphilis. Marked fluctuations are observed in this latter disease, correlated with the activity of the pathological process.

In cases under treatment the Vernes test offers an additional criterion of considerable value in estimating the progress of the condition. If the reading falls consistently one feels confidence in his therapy. Conversely a persistent high Vernes reading suggests a revision of the therapeutic regime. The test represents a distinct addition to the serological control of syphilis.

DIPHTHERIA IMMUNITY. THE EFFECT OF REPEATED INJECTIONS OF AVIRULENT DIPHTHERIA BACILLI, B. HOFMANNI AND B. XEROSIS IN GUINEA-PIGS.* M. J. Rosenau and (by invitation) G. H. Bailey, Boston.

Abstract. The object of these experiments was to determine whether avirulent diphtheria bacilli are able to stimulate antitoxin production and thus account for the acquired immunity to diphtheria which most persons develop with maturity. Seventeen different strains of avirulent cultures, and also cultures of related organisms such as *B. hofmanni* and *B. xerosis*, were injected in guinea-pigs over a long period of time. All of these guinea-pigs remained Schick-positive and when finally tested showed no immunity to diphtheria toxin. It was found that there is considerable individual variation in guinea-pigs so far as their response to injection of diphtheria toxin is concerned. Usually, guinea-pigs become Schick-negative and immune in three months. One of our animals remained Schick-positive after thirty-nine injections of small doses of diphtheria toxin (1/125 to 1/8 M.L.D.) over a period of fifteen months. This guinea-pig received a total of 1.94 M.L.D. If experiments such as ours, with avirulent cultures, could be continued over a series of years, it is possible that some immunizing effects might be manifest.

Our studies emphasize the essential difference between the so-called avirulent diphtheria bacilli and the true Klebs-Loeffler organism. Not only did the aviru-

* To be published in Archives of Dermatology and Syphilology.

* To be published in the Journal of Infectious Diseases.

lent strains fail to induce an immunity, but their agglutination reactions showed that there is a wide immunological gap separating them from the diphtheria bacillus. Some of these so-called avirulent strains have pathogenic properties. We found that a few of our guinea-pigs receiving repeated massive injections of live cultures developed disseminated focal lesions which are being studied.

No change in virulence was observed in cultures of avirulent diphtheria bacilli or *B. hofmanni* or *B. xerosis* under observation for eighteen months, and transferred for thirty-six generations on Loeffler's blood serum.

Guinea-pigs react with striking uniformity to the Schick test. All guinea-pigs appear to be like some children, in that they are persistently positive. The guinea-pig's skin seems to be more sensitive than the human skin, for a typical reaction is produced by the intradermal injection of 1/250 M.L.D. Pregnancy and general systemic infections appear to inhibit the local skin reaction.

Pseudo-reactions were not observed in our studies as a result of the injection of diphtheria toxin. But sensitized guinea-pigs responded to the injection of bacterial antigens — although this reaction is not specific. With certain bacterial extracts containing bacterial protein, a local necrotic action of the skin was produced which was interpreted as a modified Arthus' phenomenon.

Experimental confirmation was obtained of the correlation between a positive Schick test and susceptibility and a negative Schick test and immunity. In a series of guinea-pigs it was furthermore found that a definite relation exists between the weight of the animal and the M.L.D. of diphtheria toxin. It takes about twice as much diphtheria toxin to kill a guinea-pig weighing four times as much as the standard control. In other words, if the M.L.D. for a 250 gm. guinea-pig is 0.011 then it will take 0.22 for a guinea-pig weighing 1000 gm.

Morphologically the avirulent diphtheria bacillus resembles the true Klebs-Loeffler organism, but the results recorded above emphasize the fundamental difference between the two groups and throw doubt upon the appropriateness of the name "avirulent diphtheria bacilli."

FURTHER RESULTS WITH THE DICK TEST AND ACTIVE IMMUNIZATION WITH SCARLATINAL TOXIN. Abraham Zingher, New York.

Abstract. 1. The scarlatinal toxin can be standardized with only a fair degree of accuracy. Toxin prepared without blood should preferably be used to avoid confusing serum reactions.

2. Scarlet fever developed only in those who give a strongly positive Dick reaction — no case developed in a negative or slightly positive reactor.

3. The Dick test made on over 600 patients with scarlet fever showed that over 95 per cent had a slight or moderately positive reaction during the first few days of the disease and a negative reaction in convalescence. A persistent strong positive reaction in convalescence points against the diagnosis of scarlet fever. Patients admitted to the hospital as scarlet fever cases and showing such reactions should be immunized with a prophylactic dose of scarlatinal antitoxin.

4. The negative Dick reaction in the "naturally" immune is not as permanent as the negative Schick reaction; — about 10 per cent of children under 15 years of age gave a slight or moderately positive reaction when retested within a year. Among adults, however, only 1 per cent changed from a negative to a slightly positive reaction.

5. The dose of toxin for immunization has been increased, four injections, 250, 1000, 2000, 3000 skin test doses, being given at weekly intervals.

6. The rapidity in the development of immunity after scarlatinal toxin injections indicates that immunization with toxin can be used during the outbreak of scarlet fever. Care must be taken, however, under these conditions not to mistake constitutional reactions associated with rashes as clinical cases of scarlet fever. Such reactions develop in 5–10 per cent of individuals after the first dose of toxin and in only very few after the subsequent injections.

7. The antitoxic immunity after scarlatinal toxin injections develops much sooner than after diphtheria toxin-antitoxin injections. Of those who gave a negative Dick reaction at the end of one month, 20 per cent showed slight or moderately positive reactions at the end of eight to twelve months, but none had so intense a reaction as in the original test. With a second series of toxin injections complete immunity developed in these individuals and also in those who failed to be fully protected after the first series. With a sufficient number of doses of the toxin a large proportion of human beings can be successfully immunized and kept immune against the toxic effects of scarlet fever.

8. The ultimate duration of the immunity conferred by injections of scarlatinal toxin cannot be predicted at the present time. The wide distribution of the scarlatinal streptococcus makes it appear very probable that the immunity process, once successfully started, will continue in most of the persons injected as a result of contact exposure and repeated infection with the specific organism.

RAPID ALTERATIONS IN THE BLOOD SUGAR LEVEL OF RABBITS IN ANAPHYLAXIS AND FOLLOWING INJECTIONS OF BACTERIAL PROTEINS. Isolde T. Zeckwer (by invitation), Boston:

Abstract. In fatal bacterial anaphylaxis produced in rabbits, there were found to be rapid alterations in the blood sugar level. When determinations were made on blood withdrawn by heart puncture at short intervals, there occurred, after latent period, a gradually ascending curve of blood sugar, when symptoms of anaphylaxis were beginning to be manifested; then usually a relative decrease; followed by an abrupt rise to an extremely high level at the time of death, with values varying from 321 mgm. per 100 c.c. blood to 1060 mgm.

In order to determine whether this effect was due to the anaphylactic state or to the effect of the introduction of killed bacteria into the blood stream, suspensions of killed organisms of various types were injected intravenously into unsensitized rabbits and blood sugar determinations made at short intervals. In the cases of *B. coli* and *B. proteus* isolated from normal human feces, and with a stock culture of *B. paratyphosus B*, there was a gradual increase in the blood sugar level, reaching a maximum in about ninety minutes, and gradually returning to the original values in six to eight hours. The maximum values reached were 321 mgm. with *B. proteus*, 244 mgm. with *B. coli* and 208 mgm. with *B. paratyphosus B*. With *B. enteritidis* of Gaertner there was only a slight increase to 132 mgm. The increase in blood sugar with the two latter organisms is apparently in accord with the findings of Menten and Manning, who reported before this Association last year the effect on the blood sugar, of organisms of the paratyphoid B. enteritidis groups recovered from animals suffering from spontaneous infections by these organisms. Bacteria which produced no noteworthy effect upon the blood sugar level were *B. typhosus*, *B. paratyphosus A*, *B. fecalis alkaligenes*, *B. pyocyaneus*, staphylococci, hemolytic streptococci, and non-hemolytic streptococci.

The character of the curve of blood sugar in anaphylaxis and the maximum height attained were quite similar whether anaphylaxis was produced by the

injection of organisms which in themselves altered the blood sugar level in the unsensitized animal, or whether by those organisms which had no such effect. In fact, the highest value of blood sugar in anaphylaxis was produced by the injection of killed non-hemolytic streptococci, which had repeatedly been shown to have no effect on the blood sugar in the unsensitized animal.

In considering the mechanism of the production of these blood sugar changes, the phenomena cannot be due to failure in the utilization of glucose. The extreme heights of the blood sugar level attained in such short periods of time can only be accounted for by increased production of glucose through rapid mobilization of glycogen. The effect is apparently similar to the increased glycogenolysis produced by adrenalin and by stimulation of the sympathetic nerves to the liver.

BILHARZIA IN THE APPENDIX. Alfred Plaut, New York.

Abstract. The purpose of presenting this paper is to emphasize three facts: (1) the occurrence of Bilharziosis in the large cities of the East, (2) the possibility of complete absence of symptoms from intestine, (3) the fact that the position of the spine in the egg is not characteristic for a different species of bilharzia.

The patient F.T., colored, 26 years, living in New York for many years, was in the hospital with the diagnosis: Retroversion, Endocervicitis. Her complaint was profuse discharge and occasional backache. Operation was done for retroversion, the lacerated cervix was amputated; the appendix gave the impression of subacute involvement and was taken out. Nothing in the previous history or in the findings pointed toward an intestinal condition.

The gross appearance of the appendix was normal, length 7 cm., thickness normal, serosa slightly injected. The microscope shows many tubercle-like formations in all layers of the wall. They consist chiefly of swollen connective tissue cells and of some eosinophiles. In their centers the eggs of *Distoma* (*Schistosoma*) *haematobium* are found. Many of the eggs are very well preserved, in one of them a typical larval stage (Miracidium) is found, the contents of most of them indicate that they contain young developing embryos. The spines are easy to be seen in many instances, their characteristic outline can be distinguished without difficulty from shrinking and other artifacts. Some of the spines are polar, others are lateral. Eggs with polar spine and eggs with lateral spine are found nearly in the same microscopic field. Some of the destroyed eggs apparently are situated in small hyalinized blood vessels; they may have been caught there with their sharp spine. Some tissue reaction is present around all eggs found in the appendix. All transitions are found from intact eggs to complete destruction. The débris of the shell is partly surrounded by foreign body giant cells, while otherwise very few giant cells are found. Delicate onion-shell-like structures mark the place where the destruction of an ovum has been completed. The shell stains distinctly with Weigert's elastic stain. The situation of the eggs bears no relation to the direction of muscle fibers.

In the cervix, which showed papillary erosion, one calcified egg was found and one of the above mentioned onion-shell-like structures. The cervix was dissolved in antiformin but no further eggs were found.

The feces contained few eggs of bilharzia, the urine none. There was no eosinophilia in the blood. The Wassermann was positive.

The patient made an uneventful recovery and left the hospital. Her infection must date far back as the calcified egg shows. How far her abdominal organs

are the seat of bilharzia remains an open question. Laparotomy has been followed by unexpected improvement even in very severe cases of bilharziosis. *Trichocephalus dispar* was present in the feces but not in the appendix. A similar case was published from this hospital a few years ago.

PLEXIFORM NEUROMA OF THE PHARYNGEAL MUCOUS MEMBRANE. Louise H. Meeker, New York.

Abstract. A neuroma involving a whole plexus of nerves, that is, a plexiform neuroma, may occur as one of the manifestations of a general v. Recklinghausen's neurofibromatosis, or it may occur alone. In the present case a plexiform neuroma involved a large part of the plexus of the glosso-pharyngeal nerve on the right side of the throat. The patient was a girl, 14 years of age. A swelling back of the posterior pillar appeared as a fairly hard tumor-like mass, 6 cm. in diameter. There was no ulceration. Extirpation of the growth in the pharynx was somewhat difficult and nine irregular pieces of tissue were removed.

Plexiform neuroma in the pharynx has not been previously reported. In the gross specimen the translucent, vermiform nerve trunks could easily be teased out by aid of the dissecting microscope.

The striking thing in every section was the abundant convoluted nerve trunks, larger and smaller, ramifying throughout a scanty connective tissue stroma in every direction. Mucous glands and muscle bundles were invaded by the tortuous nerves.

The nerves show well-preserved medullated nerve fibers and abundant ganglion cells in clumps or embedded singly among the fibers.

Many ganglion cells have a capsule as in the sympathetic system. Certain ganglion cells and nerve fibers appear to be related to each other. The tissue immediately about the nerve fibers and ganglion cells is neurogenous tissue.

There has been much discussion as to the exact nature of the growths. They have been compared to harmatoma (Albrecht), have been considered dystrophies (Herxheimer and Roth) in contradistinction to neurofibroma (v. Recklinghausen), or true neuroma (Durante, Verocay, Askanazy, Wallner). All agree to an underlying embryonal malformation but as Herxheimer and Roth remark, "there is no agreement as to where malformation stops and tumor begins."

The plexiform neuroma in this case seems to stand in an intermediate position between the plexiform neuromas of Verocay and Herxheimer and Roth on the one hand and the ganglion neuromatosis of Krauss or Obendorfer on the other hand.

CONGENITAL SARCOMA OF THE LEG (Bone Sarcoma?). REPORT OF A CASE WITH EIGHT YEARS' CURE. Anatole Kolodny, Iowa City.

Abstract. An infant girl eight weeks old with a congenital tumor of the left leg, a Roentgenogram showing bone involvement suspicious of bone sarcoma was admitted to the Iowa State University Hospital in 1917. An amputation through the middle of the thigh followed with a permanent cure as regards a recurrence to date.

Pathology. A tumor which replaced practically all of the soft tissues of the leg, covered with normal skin, proved to be a sarcoma with a perivascular arrangement in the younger portions and a seeming differentiation into a

fibrosarcoma in the older. The relation of the tumor to the bones of the leg could not be decided from the morphology of the tumor. The very intimate relation of the tumor to the bones, especially the fibula, with the evident involvement of the bones on Roentgenogram would seem to speak for bone origin of the tumor.

RENAL NEOPLASMS IN YOUNG CHILDREN. Martha Wollstein, New York.

Abstract. Seventeen primary tumors of the kidney were studied grossly and microscopically. Six occurred in infants under one year of age, nine in children between one and three years, and two in patients three and a half and six years old respectively. All were unilateral, retroperitoneal, within the kidney capsule and compressed the kidney substance so that a layer of fibrous tissue from which all renal structures had been eliminated separated neoplasm and kidney. Any portion of the kidney may be involved. Seven were firm in consistence. Ten were soft, even fluctuating in places.

Microscopically all the growths were embryonal in type. There was one spindle cell sarcoma and two adenomata. In the other fourteen tumors embryonal epithelial elements and embryonal connective tissue elements varied in comparative quantity and arrangement, defining several sub-groups.

The less differentiated the component cells, the more rapid, soft, and invasive the growth, and the more fatal the prognosis. The more highly differentiated the cells, the firmer and less invasive is the growth, and the greater is the possibility of cure.

The more solid tumors are more easily removable, because they remain within their capsule unbroken. From the three recovered cases they were all removed entire. The more rapidly growing tumors break through their capsule and recur locally; they may also metastasize in the lungs and liver. Metastases occur through the blood stream.

UNUSUAL MODES OF DISSEMINATION OF OVARIAN CARCINOMA.* Isaac Levin and (by invitation) Joseph Barnet, New York.

Abstract. In the course of the last year the writers have observed amongst the pathological material at the New York City Cancer Institute a series of cases of ovarian carcinoma in which the secondary tumors have invaded the vital organs and caused clinical symptoms stimulating primary conditions in such organs.

In ovarian carcinoma the secondary tumors developed most frequently not through multiplication of cancer cells which were transported as an embolus through the lymph or blood circulation but by direct implantation of the cancer cells on the parietal or visceral peritoneum, or by Handley's method of lymphatic permeation.

The cases reported in this presentation showed invasion of the stomach, the body of the uterus, and the cervical endometrium. Clinically the cases simulated primary carcinoma of the stomach and carcinoma of the body of the uterus and the cervix. A detailed report of the clinical history and postmortem findings is given.

* To be published in the Archives of Clinical Cancer Research — under the auspices of the New York City Cancer Institute.

THE EFFECT OF THE SCORBUTIC STATE UPON THE PRODUCTION AND MAINTENANCE OF INTERCELLULAR SUBSTANCES. S. B. Wolbach and (by invitation) Percy Howe, Boston.

Abstract. This paper deals with the effort to determine the fundamental lesion or defect of the scorbutic state and the endeavor to make a pathological characterization of scorbutus. So far, guinea-pigs in the condition of absolute scorbutus, *i.e.*, complete deprivation of anti-scorbutic substances in the diet, have been followed, and the effects of the administration of anti-scorbutics noted. The effects have been studied in growing guinea-pigs and in the repair of lesions of bone experimentally made. The earliest effect of the scorbutic state is to be found in the incisor teeth, and is evidenced by the manner in which the formation of dentine is affected and in changes in the layer of odontoblasts. The earliest changes may be observed in six to seven days, while a few days later very striking conditions are found, among the most noticeable of which is the separation of the layer of odontoblasts from the dentine. This space is presumably filled by liquid. The odontoblasts undergo striking changes in regard to size, arrangement and staining reaction. The effect of the administration of an anti-scorbutic upon this condition is apparent within forty-eight hours and results in the prompt formation of dentine so as to fill the space caused by the separation of the layer of odontoblasts. In the bones, formation of bone ceases immediately, while osteoblasts in certain locations, particularly beneath the periosteum, continue to proliferate. This applies both to flat bones and long bones. Accumulations of osteoblasts of considerable size may occur before hemorrhages take place. That the cells under consideration are osteoblasts is proved by the effect of the administration of a single dose of an anti-scorbutic substance, because it is followed by the prompt appearance of bone matrix between the cells. A single administration of orange juice produces an effect which is easily demonstrable forty-eight hours later. In the incisions through the cortex of bone in the scorbutic state there is no bony repair, while controls operated on in the scorbutic state, but given anti-scorbutic substances after the operation, show new bone formation very promptly. The repair of soft tissues is likewise very markedly affected. Proliferation of fibroblasts is apparently but little affected by the scorbutic state, but there is a marked diminution in the amount of intercellular substance formed. There is also a very marked effect in the retardation of new blood vessel formation in the organization of lesions, etc.

Other observations were made upon the effect of the scorbutic state upon bone previously formed and upon cartilage.

The conclusions reached include the following, that the scorbutic state may be characterized as one affecting supporting tissues in which the cells are unable to produce and maintain intercellular substances. This condition affects various supporting substances to a different degree and is most marked in those in which the intercellular substance is calcified, as the dentine of teeth and the matrix of bone. The characterization applies to cartilage and connective tissue, and, by inference, to other intercellular substances, including that of blood vessels. The hypothesis is entertained, based upon the study of repair in incisor teeth of scorbutic guinea-pigs, that in the formation of intercellular substances there is a change of the material from a liquid to a solid or jell state, and that the missing factor in the scorbutic condition is one affecting the jelling or setting of a liquid product.

THE EPITHELIAL TISSUES IN EXPERIMENTAL XEROPHTHALMIA. S. B. Wolbach, and (by invitation) Percy Howe, Boston.

Abstract. This paper contains the results of the study of xerophthalmia in rats in the effort to characterize the condition pathologically. The conclusion is reached that the condition is one primarily affecting epithelia. While transitional stages may be observed, the condition of xerophthalmia results in the transformation of various epithelia into a stratified squamous keratinizing epithelium. This change has been observed in the upper respiratory tract, including the whole of the nasal passages, larynx, trachea, bronchi. In the digestive tract, the changes are most marked in the salivary glands and accessory salivary glands, and here cavities several millimeters in diameter may occur as a result of the retention of desquamated keratinized epithelial cells. Change has also been observed in the pancreas. No changes have been detected as yet in the epithelium lining the stomach and intestines. In the genito-urinary tract this transformation into keratinized epithelium is found in the renal pelvis, bladder, seminal vesicles and epididymis.

In all instances, in glands the earliest changes are to be noted in the ducts, and the histologic sequences which will be described later seem to be the same in all locations, indicating that a common factor is operative. Other changes observed include a striking atrophy of certain epithelial structures, notably thyroid gland and testes. Less striking atrophies occur in the paraocular glands and the pancreas.

The following conclusion is made: Deprivation of the anti-xerophthalmic (fat soluble A) vitamine effects specifically epithelial tissues. This effect is manifested in cells having presumably widely different chemical (secretory) functions and terminates in complete loss of specific function and in the transformation into a common type of chemically inactive (non-secretory) epithelium.

EPITHELIAL CELLS IN CONJUNCTIVAL INFECTIONS. S. Hanford McKee, Montreal.

Abstract. At the end of the third or fourth day of a gonorrhoeal ophthalmia, the clinical signs are well established. Chemosis is marked, discharge is profuse. Examination of the pus at this time may show a few gonococci but more likely no micro-organisms will be found. If now a smear be made by taking some of the epithelial cells and staining with Giemsa they will be found filled with gonococci. To the different forms of resistance met with by the gonococci as they are attempting to penetrate the epithelial cover we must add phagocytosis by the epithelial cells.

COMPENSATORY HYPERTROPHY OF THE THYMUS GLAND. J. Marmorston-Gottesman and (by invitation) H. L. Jaffe, New York.

Abstract. In the course of a series of experiments which involved the carrying out of over 100 thymectomies in rats at the age of puberty or younger, it was frequently noted that, when a small fragment of the gland was accidentally left behind at operation, autopsy disclosed a fairly large regenerated mass even as early as two weeks after the partial thymectomy. These incidental findings indicated that the gland was capable of undergoing rapid compensatory hypertrophy. But this evidence was indirect as we had no means of accurately estimating the amount of thymus left behind, and therefore a series of experiments was planned to obtain direct proof of compensatory hypertrophy.

A small number of young rats were partially thymectomized, the right lobe being completely removed; and an equal number of litter mates of corresponding sex were kept as controls. The environmental and nutritional conditions were the same for all animals. The rats were sacrificed twenty-one to twenty-four days after the beginning of this experiment; the thymus glands were removed and dissected from the surrounding fat and lymph nodes, and the individual lobes were immediately weighed. A clean dissection of the thymus from the fat and lymph nodes is a simple task in young rats.

The left lobes of the partially thymectomized animals averaged in weight 42 per cent above the left lobes of their controls; while the average difference in weight between the right and left lobes of the controls was only 5 per cent. It is significant also that the difference in body weights between the controls and operated rats was 3 per cent in favor of the controls.

These results clearly indicate that the thymus gland is capable of undergoing very rapid compensatory hypertrophy in young animals. Since hypertrophy is a response to increased physiological demands it would seem, in view of these experiments, that the organ has an important function particularly before puberty. These experiments offer further proof in support of the belief that the thymus is a gland of internal secretions.

EXPERIMENTAL ENDOMETRIOSIS. V. C. Jacobson, Albany.

Abstract. In a series of nineteen rabbits the intraperitoneal autotransplantation of endometrium during oestrus was successful in sixteen, or 84 per cent. In six rabbits so treated during the resting stage there was one positive result. In six rabbits operated on during pregnancy there were implantations in two, or 33 per cent. An increased vitality or "virulence" may be assumed for endometrial tissue during oestrus. This is in accord with Sampson's menstrual theory of origin of most cases of human endometriosis. Implantation usually occurs upon pelvic structures, and in a few instances it has been observed upon the colon and urinary bladder. The implants are invariably cystic adenomatoid structures, often multilocular. There is no evidence in these experiments in favor of the view that endometrial tissue can be formed by metaplasia of mesothelium.

CHEMICAL STUDIES ON POLYCHROME METHYLENE BLUE.* W. J. MacNeal and (by invitation) J. A. Killian, New York.

Abstract. Polychrome methylene blue is a mixture of various nearly related substances, among which methylene blue (tetra-methyl-thionin), methylene azure B (tri-methyl-thionin), methylene azure A (asymmetric di-methyl-thionin) and methylene violet of Bernthsen have been definitely identified. As a preliminary to further study of the application of these dyes in pathology the methods of preparing the dyes themselves have required attention.

Methylene blue. The medicinal methylene blue of commerce, while by no means a chemically pure substance, is satisfactory for staining work. Special brands, such as Ehrlich's rectified methylene blue, contain other dyes of this group, especially the azures. Methylene blue may be purified by recrystallization from hydrochloric acid, 20 per cent.

Methylene azure B. This substance, tri-methyl-thionin, was first recognized by Kehrmann in 1906, but the practical manufacture has been baffling. Our pres-

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ent method is as follows: Dissolve 16 gm. medicinal methylene blue in 4000 c.c. water, warm to 60° C. and add 150 c.c. of 10 per cent potassium di-chromate. Heat to 75° C., mix thoroughly and let stand in room overnight. Filter with suction. The slightly moist precipitate is transferred to a liter flask and mixed with 800 c.c. water, 5 c.c. of reagent formalin and 21 c.c. of concentrated hydrochloric acid (Sp. G. 1.19). A long reflux condenser is attached to prevent access of air and the mixture is boiled for four hours and transformed into a deep blue solution with the evolution of much formaldehyde. After very slight cooling add slowly through the condenser tube 25 c.c. of 20 per cent ammonium carbonate. Abundant carbon dioxide is evolved and the excess hydrochloric acid is neutralized. The condenser is then plugged and the mixture cooled in running water. When cold, add 350 gm. of pure sodium chloride, mix thoroughly and let stand overnight in a cool place. The precipitate is separated on suction filter, dried and recrystallized from ethyl alcohol. It finally consists of long slender needles and filaments, extremely soluble in water, from which it cannot be precipitated by hydrochloric acid. Upon addition of sodium hydroxide it becomes purplish-red and now the color may be almost completely extracted by ether, chloroform or ethylene dichloride.

Methylene azure A. Asymmetric di-methyl-thionin is made by dissolving 16 gm. medicinal methylene blue in 4000 c.c. water, adding potassium dichromate, 98 c.c. of 10 per cent solution, and 60 c.c. of concentrated hydrochloric acid (Sp. G. 1.19), and boiling for three hours. Protection from the air is unnecessary. Add 1500 gm. of pure sodium chloride, mix and let stand overnight. Recrystallize the precipitate from alcohol and dry it at 100° C.

Methylene violet. Methylene violet is the residue after removing a dimethylamine group from the methylene blue molecule and substituting an atom of oxygen for it. The reaction requires, first, oxidation and, second, hydration, in very dilute solution. Dissolve methylene blue, 8 gm. in water 5000 c.c. Add 49 c.c. of 10 per cent yellow potassium chromate and 10 c.c. of 10 per cent ammonium hydroxide. Boil for two hours. Add 15 c.c. of 30 per cent sodium carbonate and boil four hours longer. Cool and separate the precipitate on suction filter. Dry it at 37° C. and recrystallize from ethylene dichloride.

All these substances have thus been prepared in crystalline form with only moderate amounts of impurity. Their application in pathology may now be studied with greater promise of success.

THE PATHOGENESIS OF BILIARY CALCULI. S. H. Mentzer (by invitation), Rochester, Minn.

Abstract. There are two sites for the formation of biliary calculi: within the intrahepatic ducts, and in the lumen of the gallbladder. Intrahepatic stones are essentially inflammatory in origin and contain little or no cholesterin. Stones arising within the gallbladder probably can be aseptically formed and always contain varying proportions of cholesterol. Increase of cholesterol concentration of gallbladder bile may occur when the wall of the gallbladder is not capable of properly handling the cholesterol contained within it. Stasis and infection are not necessary for stone formation, but both are usually present. A nucleus of some sort is necessary for gallstone formation. A change in the acidity of gallbladder bile probably produces a precipitation of the material for stone formation.

THE PATHOLOGICAL EFFECTS OF SOLUBLE TOXIC SUBSTANCES OBTAINED FROM *B. Paratyphosus B.* Maud L. Menten, Pittsburgh.

Abstract. Soluble toxic substances were obtained from two strains of *B. paratyphosus B* (human — Brown) and PT2 isolated from rabbit and *B. enteritidis* by filtration of five-day broth cultures through Berkefeld filters and by various extractions and precipitations of suspensions of agar growths. All toxic substances recovered when injected intravenously into rabbits caused immediate rise in blood sugar reaching a maximum in two and a half to three hours and returning to normal in four to five hours. Paratyphoid toxin from animal strain gave the most marked hyperglycemia and this was followed by an eight- or ten-day interim of normal blood sugar values with a rise to about 200 mgm. per c.c. of blood, twenty-four or forty-eight hours before death.

Enteritidis toxin gave the least hyperglycemia on injection and these animals died with paralysis of hind limbs. All animals showed cellular degenerations in liver, spleen, adrenals, pancreas and kidneys. Spleen and liver were pigmented and infiltrated with laked erythrocytes. The most characteristic lesion of the paratyphoid toxin was focal necrosis of liver; chromatolysis of ganglion cells and round cell perivascular infiltration of the brain were constant when enteritidis toxin was used.

EXTRACTS OF NORMAL TISSUES IN EXPERIMENTAL TUBERCULOSIS.* Richard S. Austin, Cincinnati.

Abstract. The inoculation of saline extracts of certain organs of normal rabbits appears to influence the development of experimental tuberculosis in rabbits. The extracts of different kinds of organs vary considerably as to the amount of influence they appear to exert. Extracts of adrenal and lung would seem frequently to retard the development of lesions, those of heart and liver produce less effect, while extracts of spleen and kidney usually exert little or no influence. The freshness of the organs used, the short time utilized in preparing the extracts, and the preservation of the extracts at icebox temperature suggest the possibility that the more or less protective substances present in these organ extracts are of "native" occurrence, existing in the organs before removal from the normal animal.

LOCALIZATION OF STREPTOCOCCI FROM CASES OF EPIDEMIC NAUSEA AND VOMITING AND OF EPIDEMIC NEURO-MYELO-ENCEPHALITIS. E. C. Rosenow, Rochester, Minn.

Abstract. During the autumn and early winter of 1924, there occurred in various parts of the United States and Canada, in conjunction with relatively mild infections of the respiratory tract, an unprecedented number of cases of persistent hiccup, and cases of nausea and vomiting with or without hiccup, and of neuritis associated with or without myelitis and encephalitis. In certain communities hiccup was the most common symptom, in others nausea and vomiting, or deep-seated, unproductive coughing, while in still others neuritic pains, often unilateral, of scalp or forehead, or outspoken neuritis, dominated the picture. Marked changes in the character of the symptoms sometimes occurred during the course of the epidemic. This was true of the epidemic in Rochester. The cases of hiccup occurred chiefly between November 17 and

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December 9, 1924, and the cases of nausea and vomiting and of neuritic pains during December and the first three weeks in January.

The methods of study were similar to those used in a study of the etiology of epidemic hiccup and encephalitis. With a few exceptions, the findings in the animals paralleled in important respects those in the patients studied. The dogs injected with the material from the two patients, who were severely nauseated and vomited, developed anorexia and vomited repeatedly.

Evidence of neuritis was present in all of the five cases studied, and correspondingly, hemorrhagic lesions of nerves occurred in eight of twenty-one rabbits injected intracerebrally, and in ten of seventeen rabbits injected intravenously. Parallelism between findings in patients and animals is further shown by the fact that, of sixteen rabbits injected intracerebrally with the streptococcus from the three cases in which there was no evidence of myelitis, only one (6 per cent) had gross hemorrhages of the cord or medulla, while of twenty-two injected with material from the two patients who had undoubted symptoms of myelitis, nine (43 per cent) had gross hemorrhages in the cord.

The character of the microscopic changes in the animals injected in this series of cases was similar to that found in the experiments in epidemic hiccup and encephalitis. Hemorrhage, edema and leucocytic infiltration dominated the picture in the experiments of short duration, while later infiltration by round cells became predominant. The demonstration of organisms in the acute lesions was relatively easy; in the chronic lesions it was difficult and sometimes impossible. The streptococcus isolated in these cases is morphologically and culturally indistinguishable from the one I have isolated in epidemic hiccup and encephalitis. Like the latter, it is of relatively low general virulence. It is a slightly elongated diplococcus occurring singly and in short chains. It is gram-positive, non-encapsulated, bile-insoluble, and on blood agar produces small, dry, slightly elevated, non-adherent colonies surrounded by a green halo. All of the five strains were agglutinated by my polio-encephalitis immune serums, in dilutions as high as 1:100, and not by other similarly prepared immune serums. Positive precipitin reactions were obtained in the polio-encephalitis serums with the cleared suspensions of nasopharyngeal swabbings in three or four cases. Since changes in localizing power were noted in some of these strains following artificial cultivation and animal passage, the conclusion seems warranted that the changes in the character of the epidemic, from hiccup to other manifestations of a neuro-myelo-encephalitis, were due to change in the tropism or localizing power of the streptococcus isolated.

EXPERIMENTAL GLOMERULONEPHRITIS IN MONKEYS.* E. T. Bell and B. J. Clawson, Minneapolis.

Abstract. Intravenous injections in rabbits of several strains of streptococci from human sources produced in several instances nephritis of the spontaneous type (lymphocytic interstitial nephritis) but did not result in glomerulonephritis.

Similar injections in monkeys resulted in two cases of severe nephrosis, one of acute interstitial nephritis, one of glomerulonephritis, and one of a type not yet determined, since the animal is still living.

The case of glomerulonephritis was characterized by epithelial crescents, fusion of glomerular lobules to the capsule of Bowman, swelling of the endothe-

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lium with occlusion of the capillaries, and atrophy of tubules associated with occluded glomeruli.

Glomerulonephritis has been produced in one monkey by intravenous injections of streptococci.

EXPERIMENTAL INTRACAPILLARY GLOMERULONEPHRITIS. F. B. Mallory and Frederic Parker, Jr., Boston.

Abstract. The injection of 0.5 to 1 gm. of metallic zinc in fine powder form subcutaneously into rabbits produced acute intracapillary glomerulonephritis in four out of eleven animals killed at times varying from three to seven weeks. The time required depends apparently on the amount of local inflammatory reaction. The invading leucocytes dissolve the zinc and set it free to be absorbed into the circulation.

Proliferation of the endothelial cells lining the capillaries in the glomeruli is often active, mitotic figures are fairly numerous (occasionally two in one section of a glomerulus) and the vessels are quickly distended and occluded by the newly formed cells. The glomeruli are enlarged, containing on section two to three times as many nuclei as a normal tuft, fill the capsular space and often project into the beginning of the tubule.

The glomerular lesions are often accompanied by more or less degeneration of the renal epithelium (hyaline droplet formation, necrosis, sometimes calcification) and by regeneration of renal epithelium. The tubules contain casts and the urine albumin. Injection of zinc salts caused marked local reaction and acute tubular nephritis in a few days. Evidently slow absorption over a considerable period of time is required in order to produce the glomerular type of lesion. Two of the rabbits developed paralysis of the forelegs.

The chief source of danger to man of poisoning with zinc is from inhalation of fumes and dust in occupations involving this metal. "The chronic interstitial nephritis and paralysis from which spelter workers suffer" is interpreted by T. M. Leggs as "sequelae of plumbism" due to the frequent presence of lead in zinc. The lesions described above throw considerable doubt on this statement. Other sources of danger are foods containing acids cooked in galvanized pails, and the light fluffy stearate of zinc used as a dusting powder for babies.

Metallic nickel in powder form causes a more intense local reaction than zinc, when injected subcutaneously into rabbits, but produces somewhat more slowly the same type of proliferative changes in the vessels of the glomeruli, namely, mitoses and distension and occlusion of the capillary vessels by the newly formed endothelial cells.

THE ACTION OF GERMANIUM DIOXIDE ON THE RABBIT. C. H. Bunting, Madison, and (by invitation) G. H. Bailey and P. B. Davidson, Boston.

Abstract. Germanium dioxide in doses as small as 4 mg. per kilo has not only failed to act as a hemopoietic stimulus in the rabbit but has proved to be toxic to heart, liver and kidneys, leading to an eventual atrophy of the cellular elements of these organs.

A CONTRIBUTION TO THE KNOWLEDGE OF SPLENIC ANEMIA. Ralph G. Stillman, New York.

Abstract. A case is described in which splenectomy was performed seven years after recognition of splenomegaly. After operation hemorrhages ceased for six years and then recurred so that a second operation was performed. At the second

operation the liver was found to be normal and the splenic vein thrombosed and canalized. The normality of the liver argues for the theory that the disease is located primarily in the spleen. The occurrence and recurrence of hemorrhages can be explained on the basis of changing circulatory conditions.

INTESTINAL OBSTRUCTION AND PERNICIOUS ANEMIA. Report of a Case. Russell L. Haden, Kansas City, Kansas.

Abstract. True pernicious anemia is a clinical entity seemingly due to a specific toxic agent. The clinical evidence suggests that the toxin arises within the gastro-intestinal tract. A blood picture simulating pernicious anemia has been observed following gastrectomy, in malignant disease of the stomach or colon and with parasitic infestation of the intestinal tract. It is possible that in these various conditions there is present a common toxin which may be identical with that giving rise to true idiopathic pernicious anemia. The occasional occurrence of the clinical picture of pernicious anemia with chronic intestinal obstruction is further evidence suggestive of the gastro-intestinal origin of a hemolytic toxin. Seyderhelm noted a rapid improvement in cases of pernicious anemia after an ileostomy. Giffin and Dixon have reported similar findings. Seyderhelm has produced a megalocytic anemia in the dog by stenosing the ileum. I have recently observed a patient who presented some points of interest. A woman, 65 years old, with negative past history showed a typical blood picture of pernicious anemia. No free hydrochloric acid was present in the gastric contents. Under treatment there was marked improvement. She then developed signs of a progressive intestinal obstruction and died. At autopsy there was a marked obstruction of the duodenum and ascending colon due to a malignant growth of the gallbladder. The anemia may have been a coincident condition. It seems most probable, however, that the anemia which was clinically indistinguishable from idiopathic pernicious anemia was due to a hemolytic toxin arising in the intestinal tract. With this possibility the case is of interest in connection with the gastro-intestinal origin of the toxin of pernicious anemia.

STUDIES OF THE BLOOD IN AN EPIDEMIC OF SMALLPOX. Kano Ikeda (by invitation), Minneapolis.

Abstract. The mild pustular type shows a definite leucopenia with the initial rash, and the maximum leucocytosis during the pustular stage. Relative neutropenia is constant.

The severe pustular type shows a progressive leucocytosis with a relative neutrophilia during the initial stage and the period of desiccation. A terminal lymphocytosis occurs in fatal cases. In both pustular types the platelets are low at first but increase rapidly after the vesicular stage.

In the purpuric type there is a progressive decrease in platelets and neutrophiles with a persistent hyperleucocytosis. Normoblasts and disintegrated leucocytes are found with normal hemoglobin and red cell count.

RHEUMATIC ENDOCARDITIS AS RELATED TO SUBACUTE BACTERIAL ENDOCARDITIS. B. J. Clawson, Minneapolis.

Abstract. Eighty cases of subacute bacterial endocarditis are compared with thirty-five cases of rheumatic endocarditis in respect to (1) blood picture, (2) anatomic characteristics and (3) the bacteriology. A close pathological and etiological relationship is evident. The two types clinically are distinct because of anatomical variations. These differences seem to depend upon the character

of the inflammation (whether or not primarily proliferative), the degree of valvular involvement, and the severity and duration of the process. Rheumatic endocarditis is characterized by a proliferative type of inflammation while the subacute bacterial endocarditis is characterized by an infected thrombus on the infected valve. From these studies it seems possible that these two forms of endocarditis represent mild and severe degrees of the same infection.

ON THE INCIDENCE OF ACUTE AND SUBACUTE INFECTIVE INFLAMMATORY PROCESSES IN CARDIO-VASCULAR DEFECTS AND ON MALFORMED SEMILUNAR CUSPS. WITH A STATISTICAL REVIEW OF THE LITERATURE AND REPORTS OF SIX CASES, IN THREE OF WHICH WAS A FUNGATING MYCOTIC ENDARTERITIS OF THE PULMONARY TRUNK.* Maude E. Abbott, M.D., Philadelphia.

Abstract. Cardiac anomalies, of a form which permit the subjects to attain adult life, are very frequently the seat of a bacterial inflammatory process which is usually engrafted on a preëxisting sclerosis at the site of the defect, which marks the seat of strain. Such cases are especially malformed semilunar cusps, and septal defects and patent ductus in which an arterial-venous shunt occurs through the defect. In 555 defects "of clinical significance," acute endocarditis occurred in seventy-eight cases (17.6 per cent). The incidence was highest in malformed semilunar cusps, 18 times in forty-eight cases (45 per cent), — in defects at the upper part of the interventricular septum, 16 times in forty cases (40 per cent); in defect at the lower part of the interventricular septum (persistent ostium primum), 6 times in thirteen cases (38 per cent); in pulmonary stenosis (mostly with associated ventricular septal defect), 20 times in eighty-two cases (24 per cent); and in patent ductus arteriosus, 15 times in sixty-seven cases (22 per cent). Acute endocarditis did *not* occur in the thirty-one cases of defect of the upper part of the interauricular septum (patent foramen ovale), thus indicating that proximity to the valvular endocardium is a predisposing cause. Nor was it present in any of the cases of extreme congenital cyanosis, as these patients usually die in the first months or years of life before the age at which an acute endocarditis is liable to supervene. In septal defects and patent ductus the lesions are practically always, and sometimes exclusively, right-sided, and frequently involve the pulmonary artery and valves in a fungating growth of extraordinary luxuriance. The infection is usually of the subacute bacterial type, produced by the non-hemolytic streptococcus (Libman), but in the right-sided lesions of cardiovascular defects, acute infection by the streptococcus hemolyticus, pneumococcus, or gonococcus, is relatively commoner than in the left-sided lesions. The localization of the vegetations in the pulmonary artery and right ventricle in these cases supplies important morphological proof that the direction of the anomalous intracardiac or intra-arterial current flow is from left to right through the defect, contrary to the conclusions drawn by Holman from analogy with the condition in arterio-venous aneurysm of traumatic origin.

Six personal observations are reported, all in adults.

Case 1. Subacute bacterial endocarditis; healed and healing stages with recent exacerbation of aortic and mitral valves and margins of large defect at base of interventricular septum with dextroposition of the aorta. *Streptococcus viridans* septicemia, embolic lesions in kidneys and myocardium.

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Case 2. Perforation at upper part of interventricular septum with fused supplementary (fourth) aortic cusp and cauliflower vegetation occluding defect and protruding into right ventricle. Pneumococcus septicemia; lobar pneumonia.

Case 3. Large congenital defect at base of interventricular septum with aorta riding over it. Fungating mycotic growth completely filling pulmonary artery to its bifurcation. *Streptococcus viridans* in blood culture.

Case 4. Saccular mycotic aneurysm of the pulmonary artery at orifice of a partly obliterated ductus. Extensive vegetative endarteritis of the pulmonary artery extending down to and completely destroying the pulmonary valves. Large patent foramen ovale and hypoplasia of aorta. History of precordial pain, "bronchitis," clubbing, purpura, etc. Duration nine months.

Case 5. Patent ductus arteriosus in acute infective pulmonary endarteritis in a girl aged 19 years, dying of pneumococcus septicemia.

Case 6. Ruptured aneurysm of the right aortic sinus of Valsalva with associated interventricular septal defect and subacute infective endocarditis of margins of defect, aortic and pulmonary valves and conus of right ventricle. History of arterio-venous communication nine years, and of acute endocarditis ten months. Streptococcus septicemia.

SPONTANEOUS RUPTURE OF HEART BASED ON THIRTEEN UNPUBLISHED CASES AND SIX HUNDRED AND TWENTY FROM THE LITERATURE. E. B. Krumbhaar and C. Crowell, Philadelphia.

Abstract. Study of several cases of this rather rare condition that occurred at the Philadelphia General Hospital within a short period emphasized the fact that the usual text-book accounts are based on antiquated and inaccurate descriptions. As its dramatic qualities induce to frequent case reports and its rarity prevents its being observed often by a single individual, it seemed desirable to correlate our own findings with those reported in the literature of the past fifty years. In addition to the 13 hitherto unpublished cases that we report from the Philadelphia and Pennsylvania Hospitals and the University of Pennsylvania, we have analyzed 266 single case reports in some detail and added 354 further cases from collected studies of Quain, Minet and Le Clare and Robin and Nicolle, making a total of 633 cases, though of course no one item was studied in this number. Repetitions were avoided wherever known and other group reports were not utilized where repetition seemed likely. It can be absolutely excluded, however, only in the first two groups. Circumstances forbid giving more than the briefest outline of what has been a very time-consuming undertaking.

Spontaneous rupture of the heart is chiefly an accident to the left ventricle of the aged, and in the aged is practically always due to coronary disease. It most frequently occurs in an acute infarct of the anterior surface of the left ventricle following sudden thrombosis of an artery or a branch; or less frequently the infarct may follow gradual fibrotic occlusion of the lumen.

With severe coronary sclerosis and consequent myocardial degeneration (usually with more or less cardiac aneurysm), rupture may occur in an area not obviously necrotic and supplied by a patent artery. The bursting of a cardiac aneurysm has been observed, but is rare. Evidence is presented to show that the formerly popular diagnosis of fatty degeneration is usually incorrect or open to serious question.

Other rarer causes of spontaneous rupture considered in this analysis are "ulceration," fatty infiltration, fibrosis, syphilis, abscess, brown atrophy, parasitic cyst, tuberculosis, melanotic sarcoma. Most of these are based on reports of little value on account of the antiquity or incompleteness of the data.

Evidence about the site and character of the tear is considerable and accurate, but the actual mechanism which produces rupture and the actual cause of death are not clearly understood.

All classes and occupations are liable and the exciting causes are most diverse. Premonitory symptoms are frequent but not characteristic. Terminal symptoms are usually so abrupt that little treatment can be even attempted and the antemortem diagnosis is seldom made.